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Beekeeping together within agriculture

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ABSTRACT BOOK

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BEE BIOLOGY
09 SEPTEMBER 2019

KEYNOTE 08:30-09:30

FROM ME TO WE WITH BEES: SEARCHING FOR THE GENETIC ROOTS OF SOCIALITY
ROOM 517A

G.E. Robinson
University of Illinois, Carl R. Woese Institute for Genomic Biology, Urbana, USA

The honey bee is widely considered to be a paragon of sociality, but how did this happen? True societies are very rare in nature, but have evolved repeatedly in a group of insects that include the ants, bees and wasps. This lecture uses the honey bee and other species to show how the new science of genomics enables researchers to elucidate social life in molecular terms. We have learned that nature builds different types of social capacities in the brains of different species from common genetic building blocks, and brain systems that recognize and process stimuli that are personally rewarding can be shaped to motivate cooperation. Two additional discoveries explain how these are possible: gene activity in the brain is highly responsive to social influences, and gene regulatory networks in the brain are surprisingly malleable. These discoveries give us a new appreciation of the honey bee society.

POLLINATION AND BEE FLORA
10 SEPTEMBER 2019

KEYNOTE 08:30-09:30

INTEGRATED CROP POLLINATION IN THEORY AND PRACTICE
ROOM 517A

R. Isaacs [1], J. Gibbs [2], J. Reilly [3], R. Winfree [3], N. Williams [4], J. Wilson [1], T. Pitts-Singer [5], N. Boyle [5], M. Vaughan [6], K. Ullmann [6], K. Garbach [7], E. Lonsdorf [8], I. Koh [9], T. Ricketts [9]
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Pollination is a critical step for the production of many fruit, vegetable, and nut crops, so producers of these crops are motivated to ensure high levels of pollination. This is achieved through investment in managed bees and through the contributions of wild bees. Despite the importance of this process for yields, there is still limited information on where, when, and how best to integrate wild and managed bees into crop production systems. The Integrated Crop Pollination project has addressed these issues for major North American specialty crops, with the goal of guiding decision-making to improve sustainable pollination. In this presentation, the theory behind this project and our project goals will be reviewed along with an overview of the research conducted for multiple growing seasons on over 100 farms. Through observations of bee visits and crop production, we found that these large commercial fields are often limited by a lack of pollinators, suggesting a need for greater investment to improve pollination. Collaborators across multiple states also explored strategies for integrating pollinator habitat into these farms, developing and testing habitat for targeted species such as mason bees or to support broad bee and natural enemy populations. Evaluating these plantings for benefits to bees shows the importance of tailoring the flowering plant community to the ecology of the bees. Our research provided new insights into the role and potential of wild and alternative managed bees, but is unlikely to change grower practices without delivery of information through trusted sources. We therefore developed written, video, and web materials that have been delivered to thousands of end-users through extension networks. Understanding the social science of growers' attitudes to pollination and the sources of information was also important for this project, and our surveys have provided insights that highlight the important role beekeepers and extension professionals can play in delivering this information. This talk will conclude with a discussion of priority areas for research and extension to support effective Integrated Crop Pollination of specialty crops.

BEE HEALTH

11 SEPTEMBER 2019

KEYNOTE 08:30-09:30

WORLDWIDE PERSPECTIVE OF HONEY BEE HEALTH

ROOM 517A

P. Rosenkranz
University of Hohenheim, Apicultural State Institute, Stuttgart, GERMANY

"Honey bee health" is not only an important research field but rather has become a synonym for the state of our ecosystem and have, therefore, attained considerable public attention. According to the FAO database the number of global managed honey bee colonies has continuously increased during the past decades. This contrasts strongly with the public perception of "Bienensterben" and "CCD" and the proven periodical high colony losses in Europe and North America. Indisputable is that honey bee health is increasingly affected by certain stressors such as pathogens, availability and quality of food, exposure to pesticides, and beekeeping management. A clear assessment of these factors on honey bee health, however, is hampered by two peculiarities:
1. Honey bees exist both, as wild and managed colonies. The beekeeping business enabled the global spread of parasites and diseases. This still ongoing process leads to unbalanced host-pathogen-relationships – in the case of Varroa mites and associated bee viruses with disastrous consequences. In addition, beekeeping practices fundamentally prevent the development of natural selection towards such pathogens.
2. Honey bees live in huge entities and bee health refer not only to the individual bees but rather to the colony as a "superorganism". Pathogens and pesticides act therefore on both, the individual and colony level which complicates the determination of damage thresholds. Considering these peculiarities, examples of current threats for beekeeping will be discussed in detail, including the most important pathogens, the impact of certain pesticides and the problem of evaluating synergistic and sublethal effects. Affected by the combination of progressive globalization and intensive food production there will be an increasing risk for the worldwide distribution of pathogens and the use of new insecticides. Against this background, an outlook will focus on the requirements for future research topics and control measures in the fields of bee diseases, plant protection and agricultural biodiversity. In a final statement the importance of honey bees for the general protection of pollinators in agricultural systems will be assessed.

BEEKEEPING FOR RURAL DEVELOPMENT

12 SEPTEMBER 2019

KEYNOTE 08:30-09:30

DARWINIAN BEEKEEPING

ROOM 517A

T.D. Seeley
Department of Neurobiology & Behavior, Cornell University, Ithaca, New York, USA

Darwinian beekeeping is an evolutionary approach to beekeeping, one that seeks to provide managed honey bee colonies with living conditions that are as close as possible to those of wild honey bee colonies. The goal is to harmonize our beekeeping methods with the natural history of *Apis mellifera*, and thus allow the bees to make full use of the toolkit of adaptations that they have evolved over the last 30 million years. I will review ways in which the living conditions of honey bees differ between wild and managed colonies. I will also show how we can pursue beekeeping in a way that is centered less on treating a bee colony as a honey factory and more on nurturing the lives of honey bees.

BEEKEEPING ECONOMY

09 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

DETECTION AND PREVENTION OF HONEY FRAUD ROOM 517A

[Lead-off] Honey: What Is It And How to Ensure Its Authenticity?

S. Schwarzinger
University Bayreuth, North Bavarian NMR Centre, Bayreuth, GERMANY

Honey is probably the most natural food on the globe. Beekeepers are allowed only minimal technical intervention during harvesting and processing, while collection of the raw materials, storage in combs, mixing with bee-own substances, conversion, dehydration, ripening, and maturation must be exclusively performed by the bees. The resulting microbiologically stable, energy-rich source of nutrition for the colony has also been esteemed by humans since ages.
Today, often products containing significant portions of industrial sugar syrups are encountered in trade. These syrups closely resemble the composition of honey and can be produced from a number of raw materials (including corn, rice, wheat, …), some matching isotopic patterns in honey. Minor components suitable as markers for adulteration vary greatly as the biotechnological methods, especially the enzymes used to cleave and concert polysaccharides in these processes, as well as clean-up strategies for the syrups change. Also, drying of honey by technical means becomes a problem. Consequently, there is a multitude of syrups available and adulteration-markers, which have been identified in the past, were absent in other syrups rendering detection of adulterations difficult.
Rather than using a number of different methods for adulteration testing it would be desirable to obtain many substances in just one analytical run. This is possible with magnetic resonance (NMR) or high-resolution mass spectrometry (MS) screening. Both methods can derive markers for adulteration based on screening of large sample collections (databases) of authentic honeys and syrups that subsequently allow targeted detection of adulteration. While MS can detect very dilute substance NMR offers the advantage of being quantitative for all parameters detected, including many traditional quality parameters for honey. The high reproducibility of NMR actually allowed establishing multi-parameter ingredient fingerprints from thousands of authentic honeys from many parts in the world. This now allows targeted verification of geographic origin and variety – even in honey where pollen have been removed. In addition, knowledge of fingerprints of thousands of authentic honey samples also allows untargeted testing for unexpected or even novel deviations.
This presentation reviews the current status of developments, highlight merits of technologies but also point out needs for future developments.

Detection of adulteration by means of analysis of 2H- and 18O-isotopes in honey water

C.C. Luellmann [1], A. Duebecke [2, 3], T. Wiezorek [4], G.I. Beckh [2]
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Honey is being tested for adulteration already for several decades. Currently the only official method is the 998.12 of AOAC, which targets the ratio of 13C and 12C in the sugar and protein in honey. It works very well for additions of syrups from C4-plants like corn and cane sugar but is practically blind to additions of sugars from C3-plants, e.g. rice and beet sugar. This demonstrates the need for a method independent of the plant-source of the sugar being added to honey.
Just like honey, any syrup contains water. The isotopic composition of natural waters from lakes, rivers and sea water is already known worldwide and "isotope maps" readily available.
There is a strong correlation of 2H- and 18O-Isotopes in natural waters, which basically means that if you plot the 2H-values against the 18O-values in a coordination system, any sample of natural waters will be located on a straight line. Thus, if isotopic composition of water is shifted towards lighter isotopes, e.g. due to evaporation, the isotope values of 2H and 18 O are shifted proportionally.
Water samples extracted from honey from different locations worldwide show a different isotopic pattern, which is the basic prerequisite of this method. Additions of water to honey either directly or through the addition of syrup would shift the isotopic composition of honey water towards that of natural water samples and indicates an adulteration or possibly an illicit way of processing.
Results look very promising. The isotopic values of 2H and 18O of water extracted from authentic honeys look very different from those of natural waters. As expected, any addition of natural water or syrup prepared with natural water will cause a shift towards the isotopic composition of natural waters and thus indicate adulteration. The method is prepared for patenting process.

"Keep Honey Fraud at Bay"- Honey Authenticity Testing by 1H-NMR

L. Heintz
Bruker BioSpin GmbH, Rheinstetten, GERMANY

Food fraud is the intentional adulteration of food for financial gain, such as substitution, dilution, counterfeiting of product formulation or

packaging. According to the World Customs Organization, food fraud is costing $49 billion annually.
Food fraud is a global problem and the consequences are devastating, as the reputation of food companies, regions or countries is damaged and consumer confidence erodes.
Honey is one of the most adulterated food products and the most common frauds are the deliberate addition of sugar syrups and the false declaration of geographical and botanical origins.
As the challenges of safeguarding the global food supply chains have increased and frauds become more sophisticated, new and more advanced fraud detection procedures are needed, including new analytical methods that can fill the existing gaps.
Targeted methods for detection of sugar syrups rely on the analysis of foreign enzymes or specific markers of sugar syrups. The adjustment of the process of syrup production and purification can make such methods unable to detect adulterations.
The AOAC method 13C EA-IRMS for example is able to detect sugar syrups from C4 plants, like corn and sugar cane but is blind to sugar syrups from C3 plants, like rice, beet or wheat.
The 1H-NMR is targeted and non-targeted likewise. The technique is able to simultaneously observe hundreds of components in the honey and due to its high reproducibility acquire a so-called fingerprint of the samples. Combined with a database of authentic and known adulterated samples, the Honey-Profiling method developed by Bruker in collaboration with QSI, Alnumed and Famille Michaud allows to detect adulteration based on a multi-marker analysis.
Pollen analysis is a widely used method; however, pollen can be filtered out and pollen grains from other plants can be added to the honey to disguise the country of origin or botanical variety.
For the analysis of country of origin and botanical variety, the combination of 1H-NMR with statistical analysis represents a powerful alternative, as it relies on the chemical composition of the honey.
The method will be presented and explained in detail.

Honey Authenticity Assessment Evolution – Newly developed LC-HRMS adulteration profiling for improved detection of fraudulent addition of sugar syrups

C. Kunert, L. Elflein, K.P. Raezke
Eurofins Food Integrity Control Services GmbH, Ritterhude, GERMANY

In the last 5 years, tackling food fraud has become a major global issue. Honey is among the top ten food products which are susceptible to economically motivated adulteration. For the consumer, honey is a high-value natural product with an excellent reputation. Therefore, it is essential to safeguard the product honey with a rigid quality control using state-of-the-art methods to verify purity and authenticity. Over the last 10 years, the number of analytical methods required to detect various kinds of honey adulteration steadily increased, including stable-isotope methods, specific methods for individual adulteration marker compounds and foreign enzymes. Since a few years, 1H-NMR profiling technology has been established to provide a fast and comprehensive authenticity screening of honey, including quality parameters, geographical and botanical origin verification and adulteration detection. However, the latest experience in the routine testing of honeys from the international markets shows that there is still no universal method to detect all presently occurring sugar syrup adulterations with adequate sensitivity. Even when all existing methods, including NMR, are applied, adulterations with "honey-tailored" sugar syrups may pass the current testing regimes. Therefore, the need for an improved detection of these newly occurring adulterations is required. This was accomplished by method development and application of the LC-HRMS technology at Eurofins for which sensitive and robust instruments have become commercially available. This technique does not only allow detecting the common and newly occurring adulterations. LC-HRMS also allows combining several individual adulteration marker methods to one single multi-method detecting several hundred different adulteration markers simultaneously. Moreover, due to the combined non-targeted/targeted approach, it is also possible to identify yet unknown adulterations and newly occurring types of sugars syrups. If new adulterations will pop up in the future, the recorded data can be re-evaluated retrospectively in order to track back their first occurrence and origin. Additionally, a big advantage over other profiling methods like NMR is that no extensive database of authentic honey reference samples is required for result interpretation. As a conclusion, LC-HRMS together with stable-isotope testing (13C-EA/LC-IRMS) and 1H-NMR profiling is considered as the current gold standard for honey authenticity testing.

Technology for quality honey production in tropical regions in Latin America and Caribe

E.L. Bedascarrasbure [1], C.B. Dini [1], M.A. Palacio [2]
[1] *INTA - Centro de Investigación de Agroindustria, Buenos Aires, ARGENTINA,* [2] *INTA - EEA Balcarce, Balcarce, ARGENTINA*

First in Argentina and then in several countries of Latin America and Caribe through REDLAC, the emphasis has been placed on the technological path for quality honey production and traceability from the apiary. Production technology considers the apiary as a management unit and responds to several principles registered in PROTOCOL INTA N ° 11 (INTA PROAPI) and the Manual of Apiculture for tropical environments (www.redlac-af.org). These technologies are adapted to the conditions of each environment by the territorial technicians together with organized beekeepers and tested in the Demonstrative Units (www.redlac-af.org) where the technical and economic results can be consulted online. Although the technological path is dynamic and is adjusted to particular conditions of each region, there are some basic points as the use of standard material including honey super where the honey is ripened by dehydration by bees before cell sealing, good practices in harvesting that includes the uncapping of honey combs before extraction. More than 6000 organized beekeepers actively participate in REDLAC, but this work collects the results obtained by the Cooperativa Salvador Ferre in the Dominican Republic; ASOAPI in Costa Rica, COOPEAPI in Peru, Cooperativas Norte Grande and Flor de Garabato in Argentina that are located in subtropical and tropical environments. The results show that it is possible to obtain high quality honey in these areas without the use of technical equipment for dehydration.

Identification of Syrup Markers using High Resolution Mass Spectrometry (LC-HRMS)

G.I. Beckh [1], A. Duebecke [1, 2], T. Wiezorek [3], C.C. Luellmann [1, 4]
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The ongoing development of different modes of adulteration of honey makes it necessary to find new ways to detect adulteration of honey. There are many examples from the past that demonstrate, that a targeted approach, i.e. focusing on one or few parameters only, does only work for a short period of time until the fraudsters adapted to the new situation. Once they successfully changed the mode of adulteration, the targeted method loses its effectiveness. QSI recognized that in order to overcome this situation it is necessary to increase the number of parameter to make adulteration of honey far more complicated and less profitable for fraudsters.
To tackle this project, the latest cutting-edge technology is being employed, namely high resolution mass spectrometry. First, we analyzed a broad range of syrup and honey samples in a non-targeted way, which means that we were looking basically at any substance entering the mass spectrometer. In the next step we compared syrups with authentic honey samples by using statistical tools like principal component analysis. This analysis facilitates detection of differences in the datasets.
We identified a number of molecule masses that only occur in syrups but could not be detected in authentic honey samples (syrup marker). Thus, detection of such a syrup marker in honey is clearly indicative for the adulteration of a honey sample.
Resin treatment of honey as it is often being done in Asian countries to extract residues, e.g. of antibiotics, is another topic we focused on. We set up own resin treatments of honey in the laboratory and compared the results of LC-HRMS-analysis of honeys with that of a resin blank. The results of this test will also be discussed.

New Technologies and Business/University Collaborations in the Detection of Honey Adulteration

J. Gawenis [1], C.M. Greenlief [2], L. Gawenis [1], C. Ray [2]
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Adulteration of honey for economic benefit has evolved into multiple forms over the last 30 years. In addition to simply adding corn or cane syrup, ultrafiltration has been utilized to attempt to hide country and floral origin, aliphatic resins to remove unpalatable flavors and hide other chemical adulterants, early harvesting to artificially increase production, and bioengineered sugar syrup production to circumvent AOAC standard (C4 sugars) analyses. Recently, the development of NMR techniques in conjunction with other analytical methods has changed how adulteration testing is performed and interpreted. NMR methods now have the capacity to not only determine sugar syrup addition but also aliphatic resin use and immature harvesting. Other analysis methods can be added to further elucidate these methods of adulteration. In addition, collaboration between universities and private laboratories has created a unique opportunity to refine methods and dynamically change accreditable testing capacity in pace with the changes in adulteration methods.

BEEKEEPING ECONOMY

10 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

DIVERSIFYING INCOME SOURCES FOR BEEKEEPERS ROOM 518

[Lead-off] Pollination Markets and Bee Forage

W. Thurman
North Carolina State University, Raleigh, USA

Markets for pollination services in North America arise each spring and coordinate the activities of beekeepers and agricultural producers. After the pollination season, beekeepers migrate with their colonies to forage locations, where the economic focus turns from pollination to honey production and colony maintenance. The differences in bee stocking densities between spring and summer are dramatic. Rough calculations suggest that the colonies stocked at two colonies per acre on almonds in California each requires 80 acres of good quality forage later in the season, implying that the approximately two million colonies in the almond groves require forage area larger than the combined sizes of the states of Montana and Minnesota.
Motivated by the importance of good forage to support pollination and honey production, we discuss how contracting between beekeepers and land owners determines their economic returns and the distribution of bees across forage sites. Transaction costs of exchange, the specificity of property rights, regulatory rules, and agricultural prices all influence the functioning of this important system.

Chinese Version of "Fable of The Bees"

Y. Gao, Z. Zhao
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"Fable of The Bees" is classic paper of institutional economics and honeybee pollination contract became the typical case of externality theory. Cross-provincial transfer beekeeping accounts for 50% of the number of bee colony in China, and the annual production of honey is 65%-70% of the total. This way of beekeeping provides pollination services for China's agricultural development. Due to the uncontrollable flight range of bees, the phenomenon of hitchhiking is inevitable. The output value of nectar plant is significantly different from season, climate, kinds of pollen and pattern of beekeeping, which brings difficulties to the measurement of the value of spillover effects, even to meet agreement of pollination service. The paper uses data of fixed observed spots of national bee industry program to make statistical comparative analysis on pollination pattern, cost, and benefits, and sets up the logical framework of pollination price formation mechanism. The paper measures the coupling degree between bee and crop production based on the time of blossom, crop location, crop scale, cultivation pattern, competitive crops, beehive output and health. The results show the coupling degree of rapeseed, sunflower seed and crops grown in agricultural facilities is higher, followed by apple, citrus, tea. And those for pear and cherry is relatively lower. For crops with high coupling degree, nectar resources should be reasonably utilized and information communication channel with efficiency and convenience is necessary for beekeeper and farmer. For crops with low coupling degree, pollination service suppliers and third-party intermediary organizations will get supporting from government. More technical advocacy should be taken to promote pollination service agreement, such as grafting pollination branches, pesticide application recording on time, frequency and quantity, and promote the formation of pollination contracts.

Dynamics of queen demand and supply in Canada

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The Canadian beekeeping industry depends heavily on the importation of queens and packaged bees. In 2018, over 250,000 queens were imported, and this dependency has risks including the importation of new pathogens and the potential deterioration of queen quality from their long-distance transport. One risk factor in transport is the exposure to high and low temperatures that reduces the viability of the sperm stored in their spermathecae and alters protein expression in the queen's ovaries.
We have compared local and imported queens and observed that overall imported queens showed reduced sperm viability compared with locally produced queens. A decrease in sperm viability in queens can in turn result in decreased colony performance.
Several approaches may be considered to increase the quality of queens used in Canadian operations including a) developing methods to reduce the exposure of queens to temperature extremes during transport, b) increasing the Canadian queen supply to reduce the current dependency on imports, and c) establishing breeding selection programs to breed for stronger, disease-resistant stock.

One challenge with increasing the Canadian queen supply is the short duration of local breeding season, added to the perceived need for queens early in the year before they can be produced in Canada. This timing gap may be addressed by a modification of the beekeeping management strategy and replacing queens later in the season or by overwintering of queens or nucleus colonies to increase the local supply of queens early in the season. Results from surveying Canadian beekeepers and bee breeders, provincial apiculturists, and technology transfer team members showed beekeeper perspectives on breeding, the value of adopting new breeding tools to create stronger, more disease-resistant stock, and the desire to minimize risks associated with mass importations. A multifaceted strategy to increase the supply of local queens and reduce the risk factors for imported queens will be discussed.

Modeling the economic impact of Varroa destructor on Australian beekeepers

R. Owen, J.-P. Scheerlinck, M. Stevenson
Melbourne School of Veterinary Science, The University of Melbourne, Melbourne, AUSTRALIA

Most economic models for the impact of Varroa destructor on horticulture consider its gross cost to agriculture based on a top-down approach. It is usually assumed that beekeepers will respond to any shortage of pollination hives (due to either dissemination of wild hives or a reduction in the number of managed hives) in a timely manner. Little work has been carried out quantifying the expected costs of managing Varroa once it has become endemic and the likely impact of treatment and control on Varroa spread.
Here we model the cost of managing Varroa using a bottom-up approach. The costs for training, labour for inspections and miticide treatment including material costs were considered. The model made assumptions about expected beekeeper efficiency improvements in managing Varroa. Assumptions about annual colony losses and replacement costs for new colonies were taken into account. Labor costs were identified as the largest expense item and the time taken for each inspection/treatment could significantly affect beekeeper profitability.
The cost to prepare a new colony was determined and compared with the annual cost of Varroa treatment. As annual treatment costs per hive increase, there is a range of market values where it is cost-effective for the beekeeper not to treat for Varroa and to let colonies collapse from this disease. Indeed, at that point the replacement cost of a colony is less than the cost of treatment.
Letting a colony die allows Varroa to spread to other colonies including these owned by neighboring beekeepers, thus the cost of a beekeeper's mite infestation is partly paid-for by other beekeepers. Letting colonies die without treatment, although a rational economic choice for an individual beekeeper, is a tragedy of the commons.

Building On a Commercially Viable CIDA/Kenya Beekeeping Collaborative Program To a Transformative, Self-sustaining Enterprise

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Beekeeping in Kenya was already being practiced by some communities using traditional log hives, which produced little honey (5 Kgs per hive/year). A survey by The Kenya Freedom from Hunger and a feasibility study by Oxfam (1967-1969) proved that beekeeping could play a major role in improvement of livelihoods, particularly in rural areas. A ten-year joint project between The Government of Kenya and The Canadian International Development Agency, focused on training and extension, quality assurance and standards, hive equipment and honey refinery set- up, group and co-operative formation, for marketing of value-added products, mainly honey and beeswax. The Kenya Top Bar Hive innovation, with tremendous potential for beeswax and quality honey production (30 Kgs/hive/year), was a milestone to the enterprise. In the mid '80s, Kenya produced approximately 8000 tonnes of honey and was ranked top in Africa whereas the Co-operative Movement, which was a tool for market access, was rated second in the world (Paerpard, 2010). The industry at the time was engulfed in sustainability issues. The world Bank and The International Monetary Banks' rationalization program of '90s to curb overspending and cushion young economies from collapsing, focused on privatization and commercialization of public services, wood workshops and honey refineries inclusive, before relinquishing them to local communities. The untimely exit plans put communities at a disadvantage and resulted in ownership problems. This led to the near collapse of the enterprise. Many partners, however, came on board with tangible solutions on how to keep the initiative afloat. Inclusive were aggressive trainings, exhibitions, agricultural shows, farmer visits, media interviews to create awareness, educational visits and diversification of products. To date, there is evidence of supplementary family income derived from beekeeping, Apitherapy is embraced and the "health conscious" are enjoying a wide range of low-priced beauty products from the hive. The average honey production is currently 27,000 tonnes contributing Ksh.7.43 Billion to the GDP, with additional advantage in pollination as an ecological service of great economic value. Beekeeping is now emergent in fulfilling the Sustainable Development Goals number 1 to 3, guided by well-established Government structures and legal implements, through Public/Private/ Partnerships and collaborators.

The crisis of Ukranian beekeeping of 2018 and ways to go out from it

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[1] *Brotherhood of Ukrainian Beekeepers Ngo, Lviv, UKRAINE,* [2] *Zhen Association Llc, Kyiv, UKRAINE*

According to the data from the ITC Ukraine was one of the three largest exporters of honey in the year of 2017. Between 2011 and 2017 Ukraine demonstrated a stable growth in exports of local honey. At the end of 2017, Ukraine exported 67.786 tons to the amount of 133.684 mil.USD, which exceeded the indexes in 2011 by 6.87 times, despite the fact that import of honey to Ukraine was practically absent.
Starting from 2015 the tendencies for the future crisis in 2018 appeared. Export figures were increasing in quantitative equivalent, but at the same time the average price of exported honey began to decline substantially.
The decrease of export prices for Ukrainian honey is connected with the wrong export policy of Ukrainian exporters. Ukrainian traders chose a policy to increase Ukrainian honey exports by dumping both export and purchasing prices.
This led to a reduction in the purchase price to such a level that it became not profitable for beekeepers to produce honey.
Today the Ukrainian beekeeper can not independently export his products and is just a supplier for the trader. As a result, the tendency to reduce apiaries in Ukraine and the reorientation of beekeepers to the production of other beekeeping products began to be observed.
In order to repair this situation, the Brotherhood of Ukrainian Beekeepers NGO initiated research on the causes of the crisis, deepened cooperation with Canada and America on the issue of export of the honey bees and queen bees. What is more, a number of legislative initiatives for simplifying the business of manufacturing and further processing of beekeeping products in Ukraine were created:
• Simplified requirements for the production and further sale in the domestic and foreign markets of honey drinks were developed and approved at the legislative level;
• Requirements for beekeeping farms (producers of unprocessed beekeeping products), which will allow the beekeeper to obtain the status of an exporter;
• Amendments to existing legislation to protect bees from pesticide poisoning have been developed.
All these initiatives make the Ukrainian beekeeping industry even more attractive to attracting investment and successful business.

Diversifying Income Sources for Small Farmers through Beekeeping with Indigenous bees in India

D. Patel, S. Dhavle
Under The Mango Tree Society, Mumbai, INDIA

Under The Mango Tree Society (UTMTS), promotes beekeeping with *Indigenous bees* to increase agricultural productivity, enhance incomes and improve livelihoods of marginal farmers in India. UTMTS trains subsistence farmers in beekeeping and provides them with bee boxes which directly impact crop yields by increasing pollination. While the focus is on pollination, UTMTS also aims at diversifying income streams of beekeepers by finding a market for colonies, hive products and by developing supportive ecosystem activities that contribute to sustainable beekeeping.
Beekeepers rent out their bee colonies to bigger farmers and are remunerated in cash or kind for the pollination services rendered. In addition, new bee-friendly commercial crops are introduced to provide forage for bees in the lean season, the sale of which provide 6% additional income to smallholders. Introducing bee friendly commercial crops is extremely beneficial to farmers practicing beekeeping. Bee flora not only accelerates colony multiplication and improves the production of honey and wax, but also increases crop yield through enhanced pollination.
Sale of colonies is another route for diversification of income. There is a robust market for Cerana colonies and UTMTS facilitates the sale. UTMTS also guarantees a market for the honey produced, and 80 percent of the honey harvested is picked up at remunerative prices by its sister company that markets honey. Collection of wax and value added products with wax is the next step.
A cluster based approach for the beekeeping programme, opens up avenues for further income diversification through provision of ecosystem services. These include developing a cadre of master trainers, bee colony spotters and transfer experts from amongst the community; village carpenters for making bee boxes, womens' self - help groups for tailoring swarm bags and bee veils; and setting up bee flora nurseries for providing planting material to beekeepers.
The paper provides an overview of various income diversification measures to build a self-sustaining 'bee economy' for smallholders.

BEEKEEPING ECONOMY

11 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00
INTERNATIONAL HONEY MARKET TRENDS ROOM 518

[Lead-off] Revitalizing the Honey Market for Beekeepers

R. Phipps
CPNA International Ltd., East Norwich, NY, USA

The international honey market has reached a critical point where the integration of the incentive to produce and consume honey are in jeopardy. This stage has been reached because of the prevalence of modern sophisticated modes of illicit production and adulteration of honey.
The international market requires harmonization and development of new honey standards which are strong rather than weak, relevant rather than archaic, comprehensive rather than narrow. It also needs in the toolbox the most powerful and complementary tools for analyzing honey. Both standards and scientific tools for authenticity must fully take into account the modern modes of illicit production, including extraction of immature honey, use of resin technology, and blending of bio-engineered sweeteners. We have learned that putting a red light to some modes of adulteration and allowing a green light to others that are prevalent, will not work.
The international honey market has continued a collapse of prices, which in the view of many international experts is a consequence of the prevalence of adulterated honey created by several modes or a combination of modes. As Prof. Norberto Garcia and I have pointed out, there is a contradiction in the rising volume of honey exports worldwide and the stable number of beehives, the sharply declining productivity of the beehives and the stress and difficulty in maintaining beehive populations. Both bees and beekeepers, as Prof. Michael Roberts has expressed it, are becoming an endangered species.
We will review patterns and aberrations of prices and quantities in the international honey trade over the past two decades.
The health and vigor of the honey industry is essential to global food security and ecological sustainability. We are entering an era of enhanced traceability, increased demand for integrity in the food supply, and the creative marketing of natural products, the foundation of which is not to be found in adulteration but in an authenticity that meets the strongest, most relevant and comprehensive standards.

International honey markets facts & trends

S. Pou
Odem International Inc, Rosemere, CANADA

ODEM INTERNATIONAL INC has built a strong reputation since its foundation in 1989, dealing with numerous Canadian beekeepers and most of the honey producing countries to deliver not only Northern American markets but also Japan, EU and Gulf countries; totalizing around 8% of worldwide exchanges.
We will first present and comment various data and charts and the go for an overview on the past 19 years major facts, prior to focussing on 2 major origins growth. Finally, we will end with a more detailled study of the Canadian import market.
1.MAJOR MARKETS (Imports & Exports)
Distribution of the total exchanged volume of over 400’000 Mt of honey over 4 main import markets (Cf. chart 01).
Distribution of the same exchanged volume among various origins including Asia, Eastern Europe, South America and Northern America (Cf chart 02).
2.MAIN MARKETS FLOW OVERVIEW
A quick overview of the imported volumes per previously mentioned markets and by main origins on the past 3-5 years, including Japan, Middle East, E.U. and U.S.A. (Cf. Charts per market 03-06).
3.MAIN NOTABLE DIFFERENCES BETWEEN MARKETS
From the importing countries: consumption habits, legislation, imports origination.
From the exporting countries: Micro-Economics, weather conditions.
Substitution-ability of origins (i.e.: Argentina vs Ukraine vs India vs Vietnam).
Competition between import markets (i.e.: Argentina or Ukraine to EU vs US).
Supply & Demand.
4.FACTS & IMPACTS over the past 20 years:
Some impacting facts from the Chinese ban to EU and the Chloramphenicol crisis, to anti-dumping decisions of the US… (Cf. Chart 07).
Price levels chart since 2008 with explanations on low and high picks (Cf. Chart 08).
Example of an exporting country which grown honey production volumes 10 times over 2 decades. (Cf. Chart 09).
A word on another origin which made it through to the US lately (Cf. Chart 10).
5.THE CANADIAN MARKET STRUCTURE - a conclusion to the hosting of Apimondia
- Production and consumption (Cf. Chart 11).
- Import market per honey segment (bulk table grade, Industrial, Organic, and Retail) detailed over the past 5 years. (Chart 12-15).
- Impact of domestic crop prices and exchange rate (Chart 16).

Traceability of honey: New solutions to an old requirement

F. Vazquez [1], P. Borgna [2]
[1] *Secretaria de Agroindustria, Buenos Aires, ARGENTINA,* [2] *Senasa (Servicio Nacional de Sanidad Y Calidad Agroalimentaria, Buenos Aires, ARGENTINA*

To trace the origin of food is a mandatory legal requirement, both at national and international levels. Its main aim is to protect the health of consumers.
Traceability in the food supply chain is of critical importance to allow effective measures to be taken in the case of unsafe products. Each stage of the chain must ensure the traceability of the product. Whilst the responsibility within each company may be clearly defined, responsibility for maintaining the interfaces between primary production and the rest of the stages is often less clear.
Argentina is internationally recognized for the quality of its honey. According to FAO, Argentina is the third world honey producer (2016/2017) and the second world exporter. The country counts with more than 2,5 million geo-referenced beehives, owned by 12,000 beekeepers registered in the national register of beekeepers (RENAPA), that annually produce an average of 65,000 tons of honey, 95% of which are exported in bulk.
In that context, the Argentine government has developed a computer application whose main objective is to improve our honey traceability, thus giving improved guarantees to all our markets.
Each empty drum is now labeled with a barcode that constitutes the basis of the new traceability system. In order to ensure the full traceability of our honey, all events occurring through the production chain until the final exportation of the product must be reported to the program on line (date, process carried out, movements, etc.).
Thus, with a unique security label per drum, we know all the history of the product, implying all involved actors, such as drum manufacturers, beekeepers, honey collectors, processors, and exporters.
Our new traceability system not only ensures that Argentine companies comply with law, but also the transparency of our hole sector, the possibility of gaining new markets, the opportunity for better prices, and above all things, a greater protection of consumer´s health.

Honey fraud evidence in major importers, Australia, Japan, Usa, Spain and Uk; only solution: mandatory sampling and comprehensive analysis to all imported honey containers

F. Berron [1,2]
[1] *Amema, A.C., Merida, MEXICO,* [2] *Miel Integradora, S.A. De C.V., Merida, MEXICO*

The beekeeping industry weakens as it gravitates deeply around honey fraud. Our Istanbul presentation discussed the use of EA-LCIRMS C3/C4 authenticity analysis as benchmark for massive honey fraud, proposing as solution a world beekeeper owned NMR data base to overcome the criticism and legal dead end from the privacy of the data sources. Time elapsed has shown a key point: authenticity is to be defined by several analyses and a final expert determination, never by one sole analysis.
Following the Australian honey fraud case unveiled September 3, 2018 a similar exercise was carried in the UK.
All “honey” jars purchased at 9 different retailers tested positive for adulteration considering Psicose as the fraud marker. More than 50% showed additional fraud markers via presence of Alpha-Amylase, E-150 Color and Glycerol, all markers of different sources of adulteration. All but one sample failed the NMR testing.
Not surprisingly all passed the EA-LCIRMS C3/C4 test, confirming its use not only as benchmark for the honey fraud, but also as a fraudulent certificate of authenticity with backing of 2 major commercial laboratories in Germany. On personal conversations and a published poster, both acknowledged analysis could be tricked by a non-hive construction of its authenticity elements and/or addition of beet syrup up to 30-49%. In this way, these labs have knowingly helped to conceal the fraud: not stating in the analysis reports the true limitations of the test.
The purpose of this work is to show in details the proofs of purchase, the above mentioned analysis, concluding that despite the advance of NMR, a comprehensive analysis approach to every container imported into Europe and the USA, shall be made mandatory and performed by an independent third party. To be comprehensive, analysis shall be made to detect in each aliquot honey sample per ocean container, its markers for: biological activity, geo-botanical origin, bee activity markers, generic and targeted adulteration markers and (5) sensory tests, all examined by experts.
If such measure is not implemented shortly, the main reason for bee population collapse will be honey fraud driving the main ally of bees, beekeepers, out of business.

Blocks for Bees: Using Blockchain Technology to Ensure Honey Purity

M.A.S. Rünzel [1], B. Hadley [1, 2], E. Hassler [1, 3], J. Cazier [1, 3, 5], J. Wilkes [4, 5]
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Honey is one of the most commonly faked foods in the world. Complex value chains, limited traceability, and the lack of controls facilitate the sale and use of impure or fake honey. These days, adding sweeteners, transshipping, and mislabeling honey decreases consumers’

trust and depresses honey prices, threatening the incomes of beekeepers across the globe as it becomes harder to distinguish pure honey. Particularly, honey producers of varietal or regional honey suffer from competing, possibly fake kinds of honey. In the face of deteriorating trust in honey's authenticity, beekeepers worldwide seek ways to prove their honey's origin and quality. Emerging distributed ledger technologies (DLT) may allow restoring trust in honey.
DLTs such as the blockchain allow for the storage of information in blocks of data that are saved in a cryptographically-secure format in multiple places at the same time. Each new block of information is uniquely linked to the preceding block so that each block within the chain can only have one possible location. Hence, a blockchain provides a safe, secure and proof record of all data that is being entered. As each entry is marked with the exact time, and the chain is continuously updated, origin and production process-related information could be saved in a decentralized, unchangeable and verifiable manner. Importantly, by storing accurate and unique data about individual beehives, it is possible to remove many of the information asymmetries that exist in markets related to beekeeping. Due to its nature, blockchain technology has the potential to allow honest beekeepers to separate the market for pure honey, from the market of possibly fake honey, increasing price efficiency and largely mitigating the depressive effects adulteration has on honey prices. This also has the potential to increase price premiums by proving local origin, varietal or organic honey features. We show how a blockchain-backed system may be implemented to allow consumers to regain confidence in honey through intrinsically verifiable trust in the quality, purity, and provenance of the honey they buy. Specifically, we illustrate how blockchain technology is suitable to ensure honey authenticity through traceability from the hive to the table.

Honey trends and beekeeping in the Kyrgyz Republic

D. Muratova
Public Union Kyrgyz union of beekeepers, Karakol, KYRGYZSTAN

The Kyrgyz Republic (Kyrgyzstan) is a country of the former Soviet Union located in Central Asia. 3/4 of the territory of Kyrgyzstan is occupied by mountains up to 7439 meters above sea level. Kyrgyzstan has extensive alpine pastures that are covered with a variety of honey plants. Natural and climatic conditions favorably contribute to the development of beekeeping.
For the first time, bees were brought to territory of Kyrgyzstan to Przhevalsky district (now Issyk-Kul region, Karakol city) by Russian immigrants in the 70s of the 19th century. According with some sources bees were brought in 1878. On the south of the republic people began to be engaged in beekeeping only in the early 20th century.
Currently, there are over 1,000 beekeepers have about 50 thousand bee colonies. The main elite breeds in Kyrgyzstan are Carpathica (*Apis mellifera carpathica*) and Carnica (*Apis mellifera carnica* Pollm) and local breeding bees. According to the Ministry of Agriculture for 2017, 2 thousand tons of honey was produced. 2018 was almost 2 times less harvest due to cold weather.
Public Union "Kyrgyz Union of beekeepers" is a member of the Apimondia since 2011. After winning awards in WBA (World beekeeping awards) in Kiev, Ukraine there is increasing demand to Kyrgyz honey in the world.
Honey market of Kyrgyzstan is small. 35-40% of the produced honey is exported. In 2017, honey exported in the amount of $ 2.9 million or 1.1% of total exports of agro-industrial products or 0.1% of the world honey export. Exports of honey reached 704.1 tons. Main kind of honey to export is white honey received esparzet or sainfoin. 5-10 years ago honey exports as a raw material, but now it exports in packed. Thus, Kyrgyz honey brands appeared in foreign markets. There are also new exporting countries like China (23%), Saudi Arabia (10,5%), UAE (6%) etc.

Assuring Honey Quality in a Tropical Monsoon Climate

T.Q. Dinh
Vietnam Beekeepers Association, Hanoi, VIETNAM

Vietnam is located in a tropical monsoon climate of the South East Asia. There are six honeybee species including five indigenous species, plus *Apis mellifera*, and there is abundant vegetation for honeybees. There are about 35,000 beekeeping farmers managing ca. 1,5 million colonies. In 2018 the country produced 52,100 tons of honey, of which 43,000 tons were exported. Beekeeping in Vietnam plays a very important role in generating income, in particular for poor farmers.
Quality control in the supply chain from the beekeepers to the honey exporters is tightly regulated. Quality control systems in Vietnam like Good Beekeeping Practice, Hazard Critical Control Point (HACCP), Food Safety Plan, Traceability System, as well as True Sources, are all applied at all steps of the honey production chain including, processing, testing, packaging and shipping. EU FVO and US FDA have audited the production chain several times and approved it. Thanks to the controls mentioned above, the Vietnamese beekeeping sector has been able to flourish in spite of the disadvantages associated with beekeeping in a tropical monsoon climate (very high humidity, hot wet weather, parasites) and has managed to supply high quality honey meeting the importer country's regulations for over 35 years of continuous exporting.

BEEKEEPING ECONOMY

12 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

MARKETING, BRANDING, VALUE CHAINS-COMMERCIAL BEEKEEPERS ROOM 517D

[Lead-off] Building your Brand: The Importance of Integrity

K. Traynor
University of Maryland, College Park, College Park, MD, USA

In the age of social media, one small slip can cost a job. Ornithologist Christopher Filardi and PR specialist Justine Sacco both lost their jobs when the internet deemed them despicable. Filardi collected a rare mustached kingfisher bird as part of his research work in the Solomon Islands. The bird became an internet celebrity and Filardi lauded, until the masses realized that collected meant killed and painted Filardi a murderer. Sacco made a poorly worded attempt at humor, joking about AIDs before boarding an 11-hour flight to S. Africa. By the time she landed her offensive comment was trending as the #1 topic on twitter and her company had fired her. Integrity in your work and brand has become critical in this modern era of marketing. Creating a company brand built on a strong foundation requires honesty and transparency, qualities for which customers are often willing to pay a premium. With technological advances, we can leverage science to help build trust in our brands, offering customers a guarantee of quality. Anything can go viral. In our business, we should look at each step in the process of production and ask ourselves “If footage of this went online, would I be ashamed? Or could I stand proudly behind my product?”

A techno-economical modelling of beekeeping operations to better anticipate the season

M. Bocquet
Apimedia, Annecy, FRANCE

Current beekeeping operations are no longer based on an established and reproducible plan. From year to year dramatical changes occur in beekeeping conditions (e.g. climate adversities, amount of colony losses, market demand of nuclei, health status of the apiary). We must therefore consider a much more tactical approach of each season. However, few beekeepers practice actual anticipation of the season, considering the exercise as useless, because too hazardous.
We show that a real forecast is relevant, in order to calculate precisely the technical and economic objectives to be achieved, considering that many decisions can be taken before the season starts. Modelling the season proved to be a good tool, allowing to anticipate large number of operations, to foresee quite precisely the plan of colony renewal and honey production, and to develop a relevant dashboard to track the progress of the goals in the season. We used it with success with beekeepers of different farm size and on different climatic zones (temperate, Mediterranean and tropical).
After a global approach of the bee farm, we elaborate, with the beekeeper, a precise model of the operating system, based on the technical circuit, on the main expected honey flows and on the simulation of the growth of the colony number. We test different hypothesis or strategies for the season and take better decisions. At the end of the year, we consider the results obtained, and discuss the differences with the model. We program a new year, but as the modelling is faster, we can go further, integrating cash anticipation and calculate the time spent in each operation, what leads to a real view of the bee farm performance. The model is created individually with the beekeeper, in a coaching mode (individually or collectively) on a excel table, so each one can be rapidly autonomous in its use.
This approach is currently being used for different purposes, as a business plan tool for new commercial beekeepers, as a season anticipation aid, as a tool for organizational anticipation in collective projects, as a research tool to determine optimum operating systems.

A framework to dynamically price almond pollination contract fees through hive strength and expected yields

E. Henry [1], M.-A. Roberge [1], B. Goodrich [2]
[1] *Nectar Technologies, Montreal, CANADA,* [2] *Auburn University, Auburn, USA*

Californian almond pollination is both one of the biggest revenue streams for U.S. beekeepers and costs for almond growers. It attracts over 70% of beehives in the US, and contracting these hives represents 20% of Almond producers total costs for the season.
The effect of the strength of the hive on its capacity to pollinate more flowers and enable larger yields is not well quantified, despite the economic incentives for both the beekeeper and the grower. Hive strength, measured by number of frames with honeybees, is the common metric used for pollination contracts.
We propose a theoretical model that quantifies resulting pollination capacity for a given hive strength and its associated market contract price using datasets published by Sheesley and Poduska (1970) on the ratio of pollen (g) collected to frame count, and the ratio of winter mortality on spring time frame count from the Bee Informed Partnership Winter Loss Surveys (2015).
With this model we simulate average frame count for any given winter mortality rate to obtain a hive’s pollination capacity and its cost. We applied this model for to a theoretical almond farm which rents 1000 hives from an apiary that underwent 30% winter mortality, and we find that for with a 5% reduction in winter mortality to 25%, the almond farmer could rent 910 hives to achieve the same pollination

capacity, which represents a 8.9% reduction in hives required.
Shedding light the on relationship between a hive's strength and pollination capacity can benefit both beekeepers and almond growers. With stronger hives, beekeepers can dedicate fewer hives to almond pollination, thereby reducing labor and risks associated with hive transportation. Almond growers can spend less to obtain the same pollination output. On the macro level, the need for optimized placement of hives within orchards will play a more crucial role in the future almond orchards are expanding faster than the growth of honeybee stock.

Euromiel, working together for a better future

R. Rodríguez Monje
Euromiel, S. Coop. De 2º Grado, Mérida (badajoz), SPAIN

We are a Beekeeping Cooperative of Second Level founded in 2001 that is made up of three first level co-operatives:
Apihurdes S. Coop (Pinofranqueado). From 1992.
Montemiel S. Coop. (Fuenlabrada de Montes). From 1978.
Sierramiel S. Coop. (Torrecilla de Los Ángeles). From 1996.
Euromiel have with more than 564 beekeepers and their 349,500 associate beehives whose long tradition and exclusive right dedication to the Beekeeping activity guarantee standard stops of quality of all our products.
We transport the beehives around diferents natural areas in Spain and Portugal.
THE BEEKEEPER
Each beekeeper counts on all the average materials accredited for the extraction of beekeeping until his make available in the cooperative.
THE COOPERATIVE
It registers the given production of each beekeeper in the packages facilitated (new drums for a single use) for it, coming to the control from quality, manipulation and packaging of the product until his make available of the client-buyer.
TRACEABILITY
From our cooperative controls of handling and taking of samples are made in field to guarantee the quality of the product from their origin. At the moment of the reception of the product in our facilities one documents in addition to the origin to he himself according to the information facilitated by the own beekeeper and the verifications made in the field, being guaranteed therefore the TRACEABILITY TOTAL of the product from each beehives.
From Euromiel and our Cooperatives we work in diferents areas like:
Bee Health.
Health of the hive.
Economy for the beekeepers.
Development of our rural areas.
Fixation of population in less populated areas of Spain.
We are open to some topic that interests you.
And more things.

Experiences in introduction of the value chain approach in the beekeeping sector in the Western Balkan cross-border region of Pchinja-Krajishte

I. Djimrevska [1], B. Mohr [1], P. Markovic [2]
[1] *Deutsche Gesellschaft Für Internationale Zusammenarbeit (giz) Gmbh, Skopje, REPUBLIC of MACEDONIA – FYROM,* [2] *Regional Rural Development Standing Working Group (swg) In See, Skopje, REPUBLIC of MACEDONIA – FYROM*

The paper presents the experinces in introduction of the value chain approach in the beekeeping sector in the Western Balkan cross-border region of Pchinja-Krajishte.
The Pchinja-Krajishte cross-border region shared among North Macedonia, Serbia and Bulgaria is geographically positioned in the oldest Balkan area characterized by high biodiversity value, large areas of high mountain pastures and numerous areas with wild nature which surround the more dynamic urban areas and make an attractive region as a whole.
Although the region is facing a number of development problems, rural areas in the region have preserved the cultural heritage and tradition, being territorially determined and possessing significant natural potential, where the natural resources are not exposed to any contamination because the region has no large industrial facilities or large urban areas.
Key socio-economic feature of the region is the progressive depopulation, high poverty and high unemployment rates which puts the region in a difficult economic situation.
The predominant economic activities are agriculture and forestry, which are mainly extensive and small scale.
The agriculture sub-sector in the region, with highest up-scaling potential is beekeeping. However, the beekeeping sector in the region require collective actions to make honey value chain strengthening and diversification through adding value.
With the project:" Support to economic diversification of rural areas in Southeast Europe (SEDRA)" jointly implemented by the Deutsche Gesellschaft für Internationale Zusammenarbeit (GIZ) GmbH and Regional Rural Development Standing Working Group (SWG), the

beekeeping sector and the honey value chain is supported through different capacity development measures and targeted investments with the aim of increasing value added and improving access to finances (from public and private sources) and improvement of access to markets.
The establishment of an integrated value chain approach in the beekeeping sector in the Pchinja-Krajishte cross-border region resulted with:
•Improved effectiveness and efficiency of the beekeeping value chain;
•Creation of new job opportunities;
•Supported civil society, the public and the private sector through networking and participation in sub-regional development partnerships;
•Strengthened business relations among the beekeeping value chain actors;
•Strengthened cooperation among the stakeholders from the region;
•Promotion of the regional exchange and strengthened rural diversification.

Contractual arrangements in pollination services markets: The case of California almonds

B. Goodrich
Auburn University, Auburn, AL, USA

California is the primary supplier of almonds for world consumption, and most almond varieties are completely dependent on managed pollination services for production. Thus, a roughly $340 million pollination industry has developed, and almond pollination services contracts now make up a major portion of revenues for most commercial beekeepers in the United States.
Almond growers, pollination brokers and beekeepers often arrange pollination agreements in November or December (some even begin as early as June) of the previous year. Forward contracting is beneficial to both growers and beekeepers, because growers can contract early to lock in prices and secure colonies for pollination, and beekeepers can prepare colonies adequately to meet colony strength requirements. Additionally, many beekeepers travel to California from as far as the eastern U.S. for almond pollination, so the beekeepers need to be guaranteed a sufficiently high price to cover transportation costs before making the cross-country trip.
Aside from anecdotal evidence there is little known about the design of contractual arrangements for pollination services. Prior economic research on pollination markets has focused on the determinants of per-colony rental fees for pollination services, but these studies have largely ignored other contract provisions that may influence those prices. The objective of this research is to provide the first formal information on the nature of almond pollination agreements through a description of the results of a survey of roughly 100 almond growers conducted at the 2015 Almond Conference.
I find that nearly half of the respondents contracted with beekeepers directly for pollination, while the rest contracted with some combination of beekeepers directly and independent pollination brokers. I find that almond pollination contracts are often based on prior relationships, with 80 percent of respondents having worked with a pollination provider for more than four pollination seasons. Formal written and informal oral agreements were used to about the same extent, but this varied based on grower characteristics. The three most common clauses other than colony strength requirements used in respondents' pollination agreements regarded beekeeper access to colonies after initial colony placement, pesticide applications while colonies are in the almond orchard and late colony placement.

Presentation of the promotional activities of the Slovenian Beekeper's Association (SBA) in the field of beekeping

T. Magdic
Slovenian Beekeper'S Association, Lukovica, SLOVENIA

Beekeeping in Slovenia boasts with a centuries-long tradition, which is maintained by us beekeepers connected to the Slovenian Beekeeper's Association. The organization and connection between us beekeepers in Slovenia are certainly the keys to our successful activities.
Since the creation of the Public Advisory Service on Beekeeping in 2005, we at SBA carry out numerous projects aimed at promoting Slovenian beekeeping and its bee products. Great success is certainly represented with our own glass for honey, which is intended for Slovenian bee products.
Among our promotional activities, the event that has drawn the most attention is the promotional and educational campaign Honey breakfast, which is already being carried out for the 13th year in a row with the help of beekeepers in kindergartens and schools. I would also like to point out the project Preserve the bees through which we conduct the campaign From sowing to harvesting, which connects both beekeepers and farmers, as well as the food industry and also children.
The main objective of these promotional activities is to raise awareness among the general public about the importance conserving nature and bees and to increase the use of bee products. All these activities, through which we are constantly in contact with the public, are carried out with the aim of informing and educating the general public about the importance of preserving beekeeping and bees. All these activities are nicely rounded up by the announcement of the World Bee Day.
The results of our past work and activities are reflected in the recently changed attitude of the public towards beekeeping and the environment, more beekeeping topics appearing in the media, an increase in cultivating honey plants and in the use of chemical agents in accordance with good agricultural practice, which reduce the poisoning of bees with phytopharmaceuticals. It is also important to note that, during this time, the demand and consumption of honey as well as higher prices for honey and increased importance of Slovenian beekeeping and its products has helped develop complementing activities on farms, such as: beekeeping tourism, various honey products and apitherapy.

BEE BIOLOGY

09 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

RECENT ADVANCES IN HONEY BEE BIOLOGY ROOM 517B

[Lead-off] Bee health: from genes to landscapes

C. Grozinger
Pennsylvania State University, University Park, USA

Multiple interacting factors are driving bee declines, including parasites and pathogens, as well as biotic and abiotic features of the landscape, such as the availability of forage, pesticide use, and climate. Our studies evaluating the genomic responses of bees to multiple stressors have demonstrated that nutrition and metabolic pathways play a critical role in supporting bee health. Moreover, macronutrient ratios in pollen underpin bee resilience to diverse stressors, and bee foraging preferences are shaped by these nutritional factors. While these studies can inform pollinator forage and habitat restoration, bees can forage over large distances and thus be exposed to stressors at the landscape scale. To address these issues, we have developed online decision support tool, Beescape, to help beekeepers, land managers, growers, and policymakers evaluate the quality of their landscapes for supporting bee populations and obtain recommendations for improving their landscapes and management practices.

Evaluation of the components and functions of royal jelly from different aged honeybee larvae in queen cells

M. Feng, J. Li
Institute of Apicultural Research, CAAS, Beijing, CHINA

Royal jelly (RJ), a proteinaceous secretion of the nurse bees, is critical for determing a female larvae's developmental fate. Larva younger than 3.5 days can develop into either a queen or a worker depending on the quality and quantity of its nutrition it obtained, whereas the queen-destined larvae are exclusively fed by RJ. Knowledge on component and functional variation of RJ for the queen bee larvae during the ontogeny of instar, a critical "decision-making" period in larvae development, is largely unknown. Chemical components, anti-bacterial prosperities, proteome and N-glycoproteome of RJ obtained from queen larvae, *Apis mellifera ligustica*, at their second (48h), third (72 h) and fourth (96 h) instars were investigated. Although the contents of crude protein, 10-HDA, and acidity in RJ were varied with larval age, each component remained relative constant when evaluated in dry basis of RJ. The protein species and N-glycosylation profiles of RJ proteins were changed with larval age, demonstrating a dynamic of RJ synthesized and secreted by nurse honeybees to satisfy the age-dependent nutritional requirements of queen bee larvae. The inhibitory effect of RJ and its water-soluble fractions on some bacteria and *Paenibacillus larvae*, an etiological agent of American foulbrood, and the anti-hypertensive activity of N-glycosylated major royal jelly protein (MRJP) 1 on mice vascular smooth muscle cells were not notably influenced by RJ harvested on different instar of larvae. Membrane proteome analysis demonstrates that the killing of *Paenibacillus larvae* by N-glycosylated MRJP2 is driven by breaking of the cell wall and membrane, which inhibits respiration and chemotaxis. Furthermore, 46 novel proteins were identified that significantly expand the proteome coverage of RJ from 105 to 151. This is the first comprehensive investigation on the influence of larvae instar on RJ demanding, and adds understanding of N-glycosylated MRJP1 and MRJP2 as potential natural-based proteins in anti-hypertension and honeybee disease treatment.

Queens combat heat stress-induced loss of stored sperm viability with ATP-independent heat shock proteins

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Climate change is threatening biodiversity around the globe and one potential driver is through heat-induced reductions in fertility. All species need to reproduce to maintain viable populations, but sperm cells are highly sensitive to heat across the animal kingdom. Honey bees have viable populations on every continent except Antarctica and are also susceptible to heat stress; therefore, they could serve as environmental biomonitors for heat-induced reductions in fertility. In addition, understanding physiological mechanisms and response thresholds underlying reductions in fertility will benefit the beekeeping industry by making us better able to preserve queen health. Here, we used quantitative proteomics to investigate heat-induced changes in protein expression in honey bee ovaries, semen, and spermathecae (the queen's specialized long-term sperm storage organ). We identified 20 heat-shock proteins (HSPs) and found that only ATP-independent HSPs were upregulated in heat-shocked spermathecae, whereas no HSPs were differentially expressed in semen and one ATP-dependent HSP (HSP70) was upregulated in ovaries. The strict ATP-independence in the spermatheca, but not in semen nor ovaries, points to tightly controlled ATP usage for stored sperm. Drones (males) are mortally sensitive to heat, which may act as a quality control mechanism to ensure that queens are inseminated with high quality sperm. Finally, we analyzed spermathecae and ovaries from healthy and failing queens in the field and found that some possess a heat-shock signature, while others show divergent stress signatures in the ovaries. These signatures could serve as biomarkers for heat stress and enable post mortem diagnostics for the beekeeping industry as well as surveying the prevalence of heat-induced loss of sperm viability in diverse landscapes around the world.

In-depth Proteome of the Hypopharyngeal Glands of Honeybee Workers Reveals Highly Activated Protein and Energy Metabolism in Priming the Secretion of Royal Jelly

G.B. Tesfay [1], H. Han [1], F. Mao [2], W. Qiaohong [2], Z. Xufeng [2], W. Fan [2], F. Yu [2], H. Bin [2], M. Chuan [2], L. Jianke [2]
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Royal jelly (RJ) is a secretion of the hypopharyngeal glands (HGs) of honeybee workers. High royal jelly producing bees (RJBs), a stock of honeybees selected from Italian bees (ITBs), have developed a stronger ability to produce RJ than ITBs. However, the mechanism underpinning the high RJproducing performance in RJBs is still poorly understood. We have comprehensively characterized and compared the proteome across the life span of worker bees between the ITBs and RJBs. Our data uncover distinct molecular landscapes that regulate the gland ontogeny and activity corresponding with age-specific tasks. Nurse bees (NBs) have a well-developed acini morphology and cytoskeleton of secretory cells in HGs to prime the gland activities of RJ secretion. In RJB NBs, pathways involved in protein synthesis and energy metabolism are functionally induced to cement the enhanced RJ secretion compared with ITBs. In behavior-manipulated RJB NBs, the strongly expressed proteins implicated in protein synthesis and energy metabolism further demonstrate their critical roles in the of RJ secretion. Our findings provide a novel understanding of the mechanism
consolidating the high RJ-output in RJBs.

Cellular immunity of the honey bee (*Apis mellifera*)

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Solitary insects have developed an individual defense against parasites and pathogens. Their immune system consists of humoral and cell-mediated components. In the humoral responses antimicrobial peptides are produced and are organized into regulatory pathways. The effector cells of the cellular responses are the blood cells, so called hemocytes. The hemocytes play a key role in the cell-mediated immunity, by the phagocytosis of the microorganisms, formation of capsules around intruders which are too large to be taken up by phagocytosis, as well as by the production of matrix proteins and antimicrobial peptides. As compared to solitary insects, the immune responses of eusocial insects are complemented with communal defense mechanisms, such as hygienic behavior, grooming, hive fever, social fever and the use of antimicrobial materials for nest construction, therefore their immune system may have special elements.
In order to explore the immune mechanisms of a highly eusocial insect, we focused on the cell-mediated immune system of the honey bee (*Apis mellifera*). So far the components of honey bee cell mediated immunity - the hemocytes - were termed by their morphological and lectin binding characteristics. We have developed a discriminative typing system based on monoclonal antibodies to distinguish the main blood cell types. Three main classes of hemocytes were identified by the reactions of the antibodies: the melanizing oenocytes the phagocytic granulocytes and the plasmatocytes, which are involved in the aggregation of the hemolymph. Our antibody to plasmatocytes reacts with the Hemolectin (AmHml), while the oenocyte specific antibody reacts with the Prophenoloxidase (AmPPO).
The comparative analysis of the cellular immune reactions in the eusocial honey bee and in the solitary Drosophila revealed significant differences between the immune reactions of these two species.
Our studies may help to reveal the complexity of the immune system and the defense reactions of the honey bee and could expand our knowledge on the special immune mechanisms of solitary and eusocial insects.
Support comes from NKFI K 120140, GINOP-2.3.2-15-2016-00001, GINOP-2.3.2-15-2016-00035 and the Hungarian Beekeepers Association.

How weather affects colony growth in Canada - environment and genetics

C. Kent [1], A. Zayed [1], BeeOmics Consortium [2]
[1] *York University, Dept. of Biology, Toronto, CANADA,* [2] *Universities of British Columbia, Alberta, Manitoba, Laval, Vancouver, CANADA*

Weather strongly affects honeybee colony growth. Beekeepers know that there are challenging years and good years, but which aspects of weather are important? The BeeOmics Consortium monitored growth of over a thousand colonies from many climate zones of Canada, and analyzed the genetics of each colony. We use satellite data to characterize many aspects of weather - temperature, sun, humidity, precipitation, and wind - during the periods of colony growth. Weather accounts for 30-50% of the variation in colony growth, while genetics accounts for much of the remainder.

This talk describes which aspects of weather are most important in Canada and how colony genetics affects growth. We have results on the importance of both the queen's genetics and the genetics of her drone mates. We also relate our results to several important sources of honeybee queens for Canadians, including imported queens from Chile, New Zealand, and Hawaii, and Canadian-bred stocks that were selected for high hygienic behaviour, mite resistance, or local adaptation to a cold temperate climate.

Baby and teenager bees give their sisters grey hair

M. Eyer [1], B. Dainat [1], C. Wotzkow [2], F. Blank [2], V. Dietemann [1]
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Honeybees display a complex life history strategy, based on short living summer bees and long living winter bees. While it is generally accepted that social context of honeybee colonies affects how long workers live, there has been little research on the effects of social environment on cellular mechanisms of ageing. The yellow-brown pigment "lipofuscin" is commonly used as marker to identify cellular senescence and could be compared to humans getting grey hair earlier or later in their life. Previous research showed that honeybee workers of similar age display tissue-specific differences in the amount and size of lipofuscin (lipid granules)- accumulation, suggesting an effect of social environment on ageing. Here, we tested this hypothesis by exposing same-aged workers to different social context: presence or absence of young workers and or brood and by measuring the accumulation of lipofuscin in the hypopharyngeal gland using laser scanning microscopy. We demonstrated that social environment indeed affects cellular ageing. These findings bring new insights in the social regulation of ageing underlying honeybee's complex life history strategy.

SYMPOSIUM 13:00-15:00
HONEY BEE GENOMICS I ROOM 517B

[Lead-off] What have we learned from 2,000 honey bee genomes

A. Zayed
York University, Toronto, CANADA

The increase in the availability of individual honey bee genomes has resulted in significant progress towards understanding the evolution and adaptation of the honey bee. These efforts have identified new subspecies and evolutionary lineages and have identified a significant number of genes involved with adaptations and colony-level quantitative traits. Many studies have also developed genetic assays that are being used to monitor the movement and admixture of honey bee populations. These resources are valuable for conservation and breeding programs that seek to improve the economic value of colonies or preserve locally adapted populations and subspecies. Finally, bee genomes are being used to map out mutations that affect colony level traits. Here, I will share with you how sequencing the genomes of over 2,000 honey bee colonies or individual workers is helping us understand the evolutionary history of honey bees, and how mutations affect ecologically and economically valuable honey bee traits.

Defence response in Africanized honey bees is underpinned by complex patterns of admixture

B. Harpur [1], A. Zayed [2]
[1] *Purdue University, West Lafayette, USA,* [2] *York University, York, CANADA*

The Africanized honey bee (AHB) is among the most successful and most popularized invasive insects. It was first bred within research facilities in Brazil as a cross between local, managed European honey bees and subtropical-adapted African honey bees. Following their escape from a Brazilian research facility in 1956, AHB has become the most common honey bee population from Northern Argentina to the Southern United States. Their notoriety comes in part from their extreme defence response, whereby they can sting nest intruders hundreds of times a minute. Previous studies have explored how genetic variants contribute to differences in defence between long-separated subspecies, demonstrating it is heritable and controlled by few loci. These studies have not yet been unable to pinpoint which genes influence variation in defence response and if these genes contribute to variation within populations. We quantified defence response for 116 colonies in Brazil and performed pooled-sequencing on 30 of the most phenotypically divergent samples. We identified 63 loci containing 285 genes significantly associated with defence response. These genes have metabolic function and non-random and unexpected patterns of admixture. Our results suggest that a combination of African and West European genetics plays a role in defensive behaviour in AHBs.

Genomic Analyses Reveal Demographic History and Temperate Adaptation of the Newly Discovered Honey Bee Subspecies *Apis mellifera* sinisxinyuan

W. Shi, C. Chen, Z. Liu
Institute of Apicultural Research, Chinese Academy of Agricultural Sciences, Beijing, CHINA

Studying the genetic signatures of climate-driven selection can produce insights into local adaptation and the potential impacts of climate

change on populations. The honey bee (*Apis mellifera*) is an interesting species to study local adaptation because it originated in tropical/ subtropical climatic regions and subsequently spread into temperate regions. However, little is known about the genetic basis of its adaptation to temperate climates. Here, we resequenced the whole genomes of ten individual bees from a newly discovered population in temperate China and downloaded resequenced data from 35 individuals from other populations. We found that the new population is an undescribed subspecies in the M-lineage of *A. mellifera* (*Apis mellifera* sinisxinyuan). Analyses of population history show that long-term global temperature has strongly influenced the demographic history of A. m. sinisxinyuan and its divergence from other subspecies. Further analyses comparing temperate and tropical populations identified several candidate genes related to fat body and the Hippo signaling pathway that are potentially involved in adaptation to temperate climates. Our results provide insights into the demographic history of the newly discovered A. m. sinisxinyuan, as well as the genetic basis of adaptation of *A. mellifera* to temperate climates at the genomic level. These findings will facilitate the selective breeding of *A. mellifera* to improve the survival of overwintering colonies

Genetic Discrimination In Honeybee (*Apis mellifera*) Subspecies Inferred From Vitellogenin Gene (Vg)

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The western honeybee is widespread in Africa, Europe, and parts of Asia with a great diversity of subspecies that can be discriminated by morphometric studies. Honeybee subspecies are grouped into four evolutionary lineages: Lineage-A in central and southern Africa, lineage-C in southeastern Europe, lineage-M in northern and western Europe and lineage-O in the Middle East. Molecular markers mostly have confirmed the morphological grouping, with minor differences. Discrimination of the subspecies is important for the conservation of the honeybee biodiversity. Until today, a wide variety of molecular markers have been used to distinguish honeybee subspecies. In this study, nuclear vitellogenin gene-Vg region encoding protein that affects reproductive function, behavior, immunity, longevity, and social organization in honeybees, was selected, because of it's great importance for honeybees. To infer the genetic discrimination based on Vg in different subspecies in the four honeybee lineages, six variable Vg exon regions of new worker bee samples from *A.m. anatoliaca*, *A.m. cypria* and *A.m. meda* (O-lineage) and *A.m. litorea*, *A.m. monticola* and *A.m. scutellata* (A-lineage) were partially sequenced and these obtained sequences combined, analyzed with the published sequences of subspecies distributed in Spain, Poland, Ural region-Russia (M); southern Africa (A); Germany, Croatia, Slovenia, Egypt (C) that are available in the NCBI GenBank Database. The results of comparative analysis of Vg sequences in a previous study show that there are 26 SNP sites that differentiate M and C lineages and can be used as diagnostic markers for those lineages. As a results of the current study, two new SNP sites in Vg gene exon 2 as a diagnostic markers were found only in O-lineage. Phylogenetic construction based on new sequences and previous sequences resulted in two main groups. The first cluster consisted of two subgroups: one of the subgroups was formed by members of M-lineage, and the other consisted of members of the O-lineage and members of the C-lineage. The second cluster consisted of members of the A-lineage. The results of phylogenetic construction based on Vg, support the previous morphometric and molecular studies.

Population Genomics Provide Insights into the Evolution and Adaptation of the Eastern Honey Bee (*Apis cerana*)

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The mechanisms by which organisms adapt to variable environments are a fundamental question in evolutionary biology and are important to protect important species in response to a changing climate. An interesting candidate to study this question is the honey bee *Apis cerana*, a keystone pollinator with a wide distribution throughout a large variety of climates, that exhibits rapid dispersal. Here, we resequenced the genome of 180 *A. cerana* individuals from 18 populations throughout China. Using a population genomics approach, we observed considerable genetic variation in *A. cerana*. Patterns of genetic differentiation indicate high divergence at the subspecies level, and physical barriers rather than distance are the driving force for population divergence. Estimations of divergence time suggested that the main branches diverged between 300 and 500 Ka. Analyses of the population history revealed a substantial influence of the Earth's climate on the effective population size of *A. cerana*, as increased population sizes were observed during warmer periods. Further analyses identified candidate genes under natural selection that are potentially related to honey bee cognition, temperature adaptation, and olfactory. Based on our results, *A. cerana* may have great potential in response to climate change. Our study provides fundamental knowledge of the evolution and adaptation of *A. cerana*.

Phylogeography of the Asian Cavity-Nesting Honey Bees

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The currently recognized species of cavity-nesting honey bees are *Apis mellifera*, *A. cerana*, *A. koschevnikovi*, *A. nuluensis* and *A. nigrocincta*. All of these are Asian, with the exception of *A. mellifera*, whose natural range is primarily in Africa, Europe and the Middle East. Earlier research

using mitochondrial DNA restriction site polymorphisms and sequences revealed extensive intra-specific geographic structure in *A. mellifera* and *A. cerana*, the two species with the most extensive ranges. In Asia, the pattern of genetic variation within and among the cavity-nesting species suggested that changes in sea-level and climate associated with Pleistocene episodes of glaciation were responsible for this diversification.
Here we present the results of an analysis of relatedness among species and populations of the cavity-nesting honey bees using genomic data. We used Restriction site Associated DNA sequencing (RAD) to generate a data set of Single Nucleotide Polymorphisms (SNPs) for all of the cavity-nesting Apis except for *A. nuluensis*, as well as several populations of the dwarf bees, *A. florea* and *A. andreniformis*, and the giant bees, *A. dorsata* and *A. laboriosa*. These data were used in a phylogenetic analysis of cavity-nesting species and *A. cerana* populations from throughout its range.
We found that (1) *A. mellifera* is sister to the Asian cavity nesting species; (2) within the Asian cavity-nesters, *A. koschevnikovi* is sister to the other species; (3) *A. nigrocincta* and the "*A. cerana*" from the Philippines are mutually monophyletic sister groups (each other's closest relatives); (4) both mtDNA and genomic data show that the "black, hill and yellow, plains" *A. cerana* of India are genetically distinct and not each other's closest relatives; (5) the yellow Indian bees are a distinct lineage, sister to the *A. cerana* of Mainland Asia (including the black Indian bees) and Sundaland, which includes Indonesia, Malaysia and the southern part of Thailand; and finally (6) the Mainland and Sundaland *A. cerana* populations are each other's closest relatives.
We suggest that the "*A. cerana*" of the Philippines and the yellow Indian bees should each be recognized as distinct species, pending thorough study of their morphology, mating biology and behavior.

BeeOMICS: Towards proteomic marker-assisted selection of a range of honey bee traits

L. Foster, A. McAfee, BeeOmics Consortium
University of British Columbia, Vancouver, CANADA

Selective breeding in honey bees is challenging, in part because it is difficult to identify, measure, and rigorously select for most economically beneficial traits. Marker-assisted selection promises to make trait selection more robust and we have previously shown that proteomic signatures in worker antennae can be used to selectively breed for hygienic behaviour. The BeeOMICS project aims to extend the number of traits for which we have selection markers through a Canada-wide correlation of proteomic and genomic features with field-measured traits. In separate discovery and validation experiments over two years, we monitored nearly 1,400 colonies from British Columbia to Quebec, measuring disease-resistant traits, pathogen and mite loads, honey production, and weight gain, etc. We simultaneously collected bee samples and measured proteomic profiles of worker antennae using independent analytical methods for the discovery and validation cohorts. We will present results from the correlation of proteomic markers with features such as hygienic behaviour, defensive behaviour, grooming, honey production, and pathogen levels, including an examination of the possible functional/mechanistic links between the correlated proteins and traits.

SYMPOSIUM — 15:30-17:30
HONEY BEE GENOMICS II — ROOM 517B

Whole-genome sequencing of Honeybees from New Caledonia

D. Bailleul [1], D. Wragg [2], R. Gueyte [3], B. Basso [4], Y. Le Conte [5], K. Canale-Tabet [1], C. Costa [6], A. Gregorc [7], K. Bienefeld [8], A. Vignal [1]
[1] *GenPhySE, Université de Toulouse, INRA, INPT, INP-ENVT, Castanet Tolosan, FRANCE,* [2] *The Roslin Institute, University of Edinburgh, Midlothian, UNITED KINGDOM,* [3] *Centre de Promotion de l'Apiculture, Technopole de Nouvelle Calédonie, Bourail, FRANCE,* [4] *ITSAP, UMT PrADE, Avignon, FRANCE,* [5] *INRA, UR 406 Abeilles et Environnement, UMT PrADE, Avignon, FRANCE,* [6] *Consiglio per la Ricerca in Agricoltura e l'Analisi dell'Economia Agraria, Unità di Ricerca di Apicoltura e Bachicoltura, Bologna, ITALY,* [7] *University of Maribor, Faculty of Agriculture and Life Sciences, Maribor, SLOVENIA,* [8] *Bee Research Institute, Hohen Neuendorf, GERMANY*

Honeybee is native from Africa, Europe and western Asia and its presence elsewhere in the world is due to human imports. In New Caledonia, importation of Western-Europe M lineage honeybees from France started in the 19th century and these black honeybees were dominant until the late 1980's when an American foulbrood infestation decimated the colonies and Eastern-Europe C Lineage importation began from Australia, New Zealand and Italy. Nowadays, the hybrid honeybee is the most frequently bred but A. m. mellifera persists. The New Caledonia Islands offers the opportunity to study the genetic admixture of two human-mediated driven and temperate European subspecies into a favorable tropical landscape. Within the framework of the INRA-ITSAP SeqApiPop project, we sequenced the whole-genome of 40 honeybees sampled from Grande Terre, the largest island of the archipelago, and from 3 medium size islands (Lifou, Maré and Ouvéa). When compared with population samples from Europe including A. m. mellifera, A. m. ligustica, A. m. carnica and A. m. caucasica, the New Caledonia honeybee population indeed reveals a mixed composition of A. m. mellifera and A. m. ligustica. The honeybees from Grande Terre have a higher proportion of A. m. ligustica background whereas the honeybees from Lifou have a higher proportion of A. m. mellifera. Maré shows an intermediate profile with a higher proportion of A. m. mellifera background than Grande Terre. The Ouvéa honeybee's background is similar to Grande Terre, and yet the Ouvéa colonies originate from a recent importation of A. m. ligustica queens. Despite a substantial importation of A. m. ligustica and the decline of the first introduced black honeybee, the genetic composition of New Caledonia honeybees is still characterized by a strong presence of an A. m. mellifera background. Our whole-genome results comfort previous studies on Lifou, suggesting this island could be considered as a black honeybee conservatory in the South Pacific Ocean.

Genetic variation of Nosema ceranae populations across the globe

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Nosemosis is an intestinal disease of adult honey bees, *Apis mellifera*, that is caused by two species of microsporidia, Nosema apis and Nosema ceranae. Nosema ceranae was first detected in Asian honey bees, *Apis cerana*, and was recently detected in *A. mellifera*. It has been hypothesized that N. ceranae was originally limited to *A. cerana* in East Asia and then spread worldwide to *A. mellifera* acting as an invasive species. Populations of invasive species generally have highly reduced genetic variation, typically due to a Founder's effect. To test this hypothesis, *A. mellifera* infected with N. ceranae were sampled from Vietnam, which is believed to be close to the presumed origin of the species, and Iran, which is also in Asia but outside of the natural range of *A. cerana*, as well as in Canada, United States, Mexico and Argentina, which are located outside the natural range of *A. cerana* and *A. mellifera*. To determine the genetic variation of N. ceranae populations, SNPs in the translation elongation factor-1 alpha gene and SSRs were evaluated. SNP analysis revealed that the highest genetic variation was detected in Vietnam with six SNP types, and the most common SNP type in Vietnam was also found in all the other locations sampled, which all had identical sequences. The SSR markers showed the most polymorphisms with five SSR types in Vietnam. Variation in the SSR types from all other locations showed that N. ceranae populations were similar between Iran and Alberta, Ontario and New York, and Mexico and Argentina, which may reflect movement of *A. mellifera* queens. These results are consistent with N. ceranae being an invasive species with one or a few genotypes spreading from Southeast Asia to other parts of the world likely related to the commercial movement of honey bees.

Genomic sequencing and analysis of sacbrood viruses from *Apis cerana* and *Apis mellifera* in Taiwan

J.C. Chang [1], Y.S. Nai [2]
[1] *Department of Biotechnology and Animal Science, National Ilan University, ILAN, TAIWAN,* [2] *Department of Entomology, National Chung-Hsing University, Taichung, TAIWAN*

Apis cerana sacbrood virus (AcSBV) have been recorded in Taiwan and causes serious loss and epizootic in *Apis cerana*, since the first detection at 2015. Sacbrood virus (SBV) is a single-strand small RNA virus and belongs to the family of Iflaviridae. It causes different symptoms in *Apis cerana* and *A. mellifera*. In most of the cases, SBV usually causes serious damage to *Apis cerana*e. However, the genomic data is lacking in both SBV in either *A. cerana*e and *A. mellifera* in Taiwan. Therefore, the whole genomic sequences of *A. cerana* sacbrood virus (AcSBV) and *A. mellifera* sacbrood virus (AmSBV) from *A. cerana*e and *A. mellifera* in Taiwan were sequenced and analysis. The full length of the whole genomes are 8,578 and 8,885 for AcSBV and AmSBV, respectively. Both of two viruses encoded one polyprotein (2315 a.a. in AcSBV and 2859 a.a. in AmSBV). Moreover, the region of poly-protein sequences from these two viruses were further compared to other 10 close related AcSBVs and AmSBVs. From the result, the helicase region is conserved among selected SBVs; for AcSBV-TW, it showed 100% identical to those of CSBV-FZ, AcSBV-Viet1, AmSBV -Viet4 and AcSBV-Kor19; while AmSBV-TW showed 100% identical to those of SBV-UK, AmSBV-Viet6 and AcSBV-IndTN1. Moreover, the comparison between AcSBV and AmSBV was also performed; we found that the AcSBV had a 17-amino-acid deletion in VP1 region compared to that of AmSBV. Besides, there are other specific characters in the genomic sequences between AcSBV and AmSBV. The phylogenetic analysis based on whole genome sequences and other conserved protein regions among the SBV isolates indicated that AcSBV-TW and AmSBV-TW are close related to CSBV-FZ and AmSBV-Viet6, respectively. Genomic difference between AcSBV-TW and AmSBV-TW could provide more evidence to the different susceptibility to their host and distinguish of the AcSBV and AmSBV in field prevalence.

Characterisation of the genotype of honey bee colonies from pool sequences

S. Eynard [1], C. Sann [2], F. Mondet [3,4], B. Basso [5], K. Canale-Tabet [1], R. Mahla [2], O. Bouchez [6], Y. Le Conte [3,4], Y. Poquet [2], F. Phocas [7], F. Guillaume [8], A. Decourtye [4,5], L. Genestout [2,8], A. Vignal [1], B. Servin [1]
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Characterising the genetic diversity of populations allows to better understand their demographic history and their adaptation to selective pressures. In honey bees, this characterisation is facilitated by a relatively small genome size, but is hindered by the fact that often the unit of observation and sampling is the colony rather than a single individual. Moreover, performing large scale genetic analyses of honey bees is a real challenge, due to the specific reproduction mechanism including multi-male insemination, making the genotype of a bee colony a mixture of contribution from the queen and the mating drones. In this work we propose an approach to characterise the genotype of a colony based on pool sequencing of worker bees. We introduce statistical models for the analysis of pool sequence data allowing to reconstruct jointly individual queen genotypes of colonies and allele frequencies in bee populations. We demonstrate the performance of our approach using data on 1500 colonies collected throughout three years within the FranceAgriMer funded, BeeStrong project. Population admixture, in terms of queen sub-species composition, validation was accomplished using information on geographical and sociological organisation of the beekeepers. In addition to a better understanding of the population dynamics of honey bees, our approach

to genotyping bee colonies promises to facilitate the genetic analysis of complex traits, and can be used for genome wide association studies on phenotypes of interests, for instance to assess Varroa resistance in honey bee populations.

Contribution to the characterization of the genetic diversity of the honeybee *Apis mellifera*: case of the sex determination locus csd in European and Algerian honeybees

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Sex determination in the honeybee is under the control of the csd gene composed of 9 exons, the seventh of which being hypervariable. A bee will be a female worker or a queen only if it is heterozygote at this specific locus and individuals having a single allele will develop as males, which is typically the case of the normal haploid males. Diploid individuals, homozygotes for the locus will also develop as male larvae, that will be killed by the workers.

We focused here on the diversity at csd by direct Sanger sequencing of PCR products of the hypervariable exon from haploid males. We analyzed a total of 257 individuals from the 5 subspecies A. m. mellifera (n=86), A. m. ligustica (n=29), A. m. carnica (n=37), A. m. intermissa (n=35) and A. m. sahariensis (n=39), in addition to bees from French beekeepers (n=51), which are a mixed type.

More than 80 DNA and 70 amino acid haplotypes were observed, many of which had not been described to date. Polymorphisms include a variation of length of a short tandem repeat-like sequence, causing the length of csd exon 7 to varie between 73 and 101 amino acids, small indels and SNPs. Most mutations observed at the DNA level translate into amino acid changes. Across all the studied population, the mean number of individuals in which one specific amino acid haplotype is detected is 2.5 ± 2.1.

When excluding the 51 mixed type bees, allele sharing between different bee types was higher than expected. For example, in the European bees, amongst 33 amino acid haplotypes observed in at least two individuals, 8 (24 %) could be detected in both M-type and C-type samples. Such a high level of allele sharing between otherwise genetically distant sub-species is surprising. This suggests either pressure for high allele diversity acting against loss by drift which could be due to the sex determination mechanism, or the convergent appearance of alleles in different populations due to a high mutation and fixation rate.

Genetic Diversity in an admixed expat Population of Honeybees in New Zealand

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Lack of genetic diversity and the resulting inbreeding effects can be a substantial problem in bottlenecked populations, especially in species with a low parent / offspring ratio where only small numbers of parents are required to repopulate the entire population. Previous work has shown that these issues are present in the North American honeybee population (Delaney, 2008; Harpur et al., 2015), leading to a push for importation of Old World genetics into the United States. Due to high levels of biosecurity and low initial numbers of settler (with accompanying bee hives), the New Zealand population can be expected to show a similar bottlenecking or founder effect. With recent changes in industry structure where large (mega commercial) beekeeping enterprises hold almost half of all hives (in June 2017, 37 operators held 38%, out of a total of close to 8,000 registered beekeeping operations), it is feared that bottlenecking between generations (a natural side effect of queen selection which is usually overcome by free mating) is becoming disproportionate. In order to satisfy growing concerns that honeybee importation laws might have to be changed in order to maintain a healthy population, we investigated the level of genetic diversity present in the New Zealand honeybee population as well the potential presence of population structure. We collected and analysed worker samples from around 25 locations throughout the country, with a focus on both hives held by large commercial operators and queen producers. Samples were analysed using Genotyping-by-Sequencing, a reduced representation genotyping method that we had previously used to genotype queens, and a high-throughput typing method for the sex-determination locus csd. While we found significant homogenisation (i.e. lack of genetic diversity) in parts of the population, the number of csd alleles present in the country was found to be sufficient to maintain a healthy population. Tools for the visualisation of the distribution of populations clusters are being developed. These will enable beekeepers to make more informed decisions about queen movements within operations and / or purchases.

De novo genome assembly of a western European *Apis mellifera mellifera* black bee

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The honeybee reference genome assembly Amel4.5, recently updated under the name Amel_HAv3.1, was produced from a commercial strain whose genetic makeup is different from the original western European *Apis mellifera mellifera* black bee subspecies, still used by some beekeepers on the continent. To construct a black bee genome assembly, we used the Pacific Biosciences long-read technology

and produced all sequence reads from a single haploid drone in order to avoid assembly artifacts due to polymorphism. To ensure we sequenced a stable representative of the subspecies originally present in western Europe, this drone was sampled from a black bee conservatory established more than 30 years ago on the small island of Ouessant in Brittany, France and since then maintained closed at the level of the island.

A total of 200 contigs (gap-free sequence tracts) were obtained. The longest contig is 11.6 Mb and the N50 contig size is 5.1 Mb. This is a great improvement in comparison to the 46 kb N50 contig of Amel4.5. To order and orient our contigs along the chromosomes, we used published sequencing reads to build a genetic map. This map is 50 Morgans long and the average recombination rate in the genome is 23 cM/Mb, which is closer to the first estimations based on the microsatellite genetic map, than to the most recent ones based on SNPs derived from sequence data. Alignment of our assembly to Amel4.5 showed agreement on the chromosomal assignment of contigs but revealed many differences in the internal ordering of large chromosome segments and typically, recombination hotspots detected on Amel4.5 at the breakpoint positions between the two assemblies, disappear when the sequence order and orientation are corrected as in our assembly. Moreover, our assembly is in very good agreement with the new Amel_HAv3.1 reference. This new reference will allow detailed analyses of structural rearrangements, possibly involving differences in gene composition, between the genomes of C-type or hybrid honeybees used by the majority of beekeepers and the M-type subspecies A. m. mellifera black bee.

A novel method to identify sex alleles of the honey bee

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More than fifty years before discovering of sex chromosomes, it was revealed that unfertilized eggs of the honey bee develop into haploid drones and fertilized eggs develop into diploid females.

However, it was uncovered only in the beginning of the 21st century, that not haplodiploidy alone, but an autosomal locus with cascades of sex determining genes is responsible for sex determination. In the honey bee, gene csd represents the primary signal that directs sexual development through the actions of alleles.

The mechanism is called complementary sex-determination where homozygosity in the sex alleles results a male - irrespectively of the ploidy level. These homozygous diploid drones are lethal: they are eaten by the workers shortly after emerging from eggs, resulting in shotgun brood as a sign of inbreeding. Natural mechanisms, as queens' avoid nuptial flight in the vicinity of her colony, their mating with multiple drones, or existence of the drone congregation areas all serves for diversity of alleles in the superorganism of the hive.

On the other hand, labour intensive closed breeding programs (eg. for Varroa tolerance) lack these evolutional assurances and diploid drones may appear quickly. Thus, inbreeding remains a big threat without being aware of the csd alleles especially when single drone insemination is applied.

The objective of our work was to provide a novel method to detect allelic variants of the csd gene directly from the queen. It enables the detection of her csd variants before insemination, and can be used to gain genetic information from the drones as well.

In closed breeding programs it can provide an alternative for labour extensive traditional approaches, like molecular cloning and subsequent sanger dideoxy sequencing or genotyping of drones reconstructing the queen.

Furthermore, we demonstrated the applicability of our method on honey samples, providing a tool that can help to determine the origin of honey samples via uncovering the sex allele composition or determining the frequency of the available alleles in a mating yard.

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BEE BIOLOGY

10 SEPTEMBER 2019

SYMPOSIUM 13:00-15:00
BEE SEMIOCHEMICALS ROOM 518

[Lead-off] Honeybee chemical ecology and interactions with stresses involved in colony losses

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Chemical communication is a fascinating aspect of social insect biology and the honeybee is probably one of the most extensively studied model in chemical ecology. Honeybees use pheromones to communicate between individuals and to regulate the development and social interactions of the colony as well. More than 50 chemical compounds have been identified having pheromonal effects and honeybee chemical ecology can be described as complexity, synergy, and context dependency, mediated through both temporal and spatial distribution.
Massive honeybee losses have been reported in many places in the world since few decades. There are evidences for interactions between stresses related to honeybee losses and chemical communication processes, modulating production or reception of pheromonal compounds. Moreover, there are examples of honeybee pheromonal compounds being used by both the host and its parasites. For example, Varroa destructor can disrupt chemical communication between individuals of the colony, but the mite can also recognize its host using its pheromone, and it can be recognized and destroyed by worker bees using semiochemicals produced or induced by the mite. I will present a review of the findings on pheromones produced by the honeybee colony giving a special emphasis on the different primer pheromones and their interactions on social regulations between the different actors of the colony, as well as their central place in the host pathogens relationships.
Finally, the major challenges for future research in the field of chemical communication in the honeybee will be presented for discussion.

Olfactory eavesdropping of predator alarm pheromone by sympatric but not allopatric prey

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Eavesdropping is predicted to evolve between sympatric, but not allopatric, predator and prey. The evolutionary arms race between Asian honey bees and their hornet predators has led to a remarkable defence, heat-balling, which suffocates hornets with heat and carbon dioxide. We show that the sympatric Asian species, *Apis cerana* (Ac), formed heat balls in response to Ac and hornet (Vespa velutina) alarm pheromones, demonstrating eavesdropping. The allopatric species, *Apis mellifera* (Am), only weakly responded to a live hornet and not to hornet or Ac alarm pheromones. We observed typical hornet alarm pheromone releasing behaviour, hornet sting extension, when guard bees initially attacked. Once heat balls were formed, guards released honey bee sting alarm pheromones: isopentyl acetate, octyl acetate, (E)-2-decen-1-yl acetate, and benzyl acetate. Only Ac heat-balled in response to realistic bee alarm pheromone component levels, <1 bee-equivalent (1 µg), of isopentyl acetate. Detailed eavesdropping experiments showed that Ac, but not Am, formed heat-balls in response to a synthetic blend of hornet alarm pheromone. Only Ac antennae showed strong, consistent responses to hornet alarm pheromone compounds and venom volatiles. These data provide the first evidence that the sympatric Ac, but not the allopatric Am, can eavesdrop upon hornet alarm pheromone and uses this information, in addition to bee alarm pheromone, to heat-ball hornets. Evolution has likely given Ac this eavesdropping ability, an adaptation that the allopatric Am does not possess.

Variation in worker behavior towards queens during colony introduction

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In the face of widespread pollinator declines, including high annual colony losses of honey bees (*Apis mellifera*), beekeepers have increasingly turned to commercially produced queens from stocks that have been selectively bred for high quality, with over one million of these queens purchased each year in the United States. However, these efforts and expenses are wasted when colonies fail to accept these queens, either by rejecting them immediately upon introduction or by superseding new queens shortly after establishment. While there is ample research on the determinants of queen quality, there is more to be known about foreign queen acceptance, particularly the worker-queen interactions that occur during introduction. We investigated queen acceptance across five different commercial queen stocks by introducing them into Italian worker populations, a stock that is commonly used in the U.S. We observed worker-queen interactions over a 10-day period to better understand the level at which the collective decision of queen acceptance is made in honey bee colonies and the specific cues involved. We found that there are no significant differences among the specific honey bee lines and rejection rates, but roughly one-quarter of the queens were not accepted by the resident workers. We will be analyzing interactions among the different lines, retinue numbers, and other observed behaviors. In addition, we will map both the genetic distances and variation in Queen Mandibular Pheromone (QMP) to determine how genetic relatedness and queen pheromonal signals interact with the decision of honey bee workers to accept or reject an introduced queen. This is a requisite first step in understanding honey bee biology and behavior during queen acceptance, while simultaneously addressing an important knowledge gap in successful apicultural management.

Sensory bias for alloparental care drives reproductive investment of honey

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The timing and amplitude of reproductive effort are central life history variables for all organisms. In social insects, reproductive effort is collectively controlled at the colony level but little is known about the mechanisms that determine how much colonies invest in reproduction. As part of their female reproductive investment, honey bee colonies raise multiple new queens by feeding royal jelly to female larvae. Artificial selection for commercial royal jelly production in China has generated over the past 40 years a stock of royal jelly bees that raises an order of magnitude more queens and provisions each queen with >3x more royal jelly than unselected stock. Here we establish in a reciprocal cross-fostering experiment that this dramatic shift in social phenotype is due to changes in the nurse bees that care for the brood. We demonstrate higher electrophysiological responsiveness to brood pheromones in royal jelly bees than in unselected bees. Comparing the antennal proteome of unselected and royal jelly bees, we identify proteins involved in chemosensation and energy metabolism as candidates for the observed differences. We confirm several candidates, most prominently OBP16 and CSP4, with quantitative differences of corresponding mRNA levels and functional binding assays between the brood pheromones and the chemosensory proteins. Furthermore, we complement analyses of brood volatiles and electrophysiological recordings with behavioral attraction assays to confirm the presumed biological function of one newly discovered and two existing larval pheromones. Together, these findings help our understanding of pheromonal communication in honey bees and explain how sensory changes in nurse honey bees as alloparental caregivers have evolved in response to artificial selection, leading to a profound shift in colony-level resource allocation to sexual reproduction.

Chemical signals of *Apis mellifera iberiensis*: glandular pheromones versus in vivo emission of volatiles compounds

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The communication in social insets, particularly honeybees, is regulated by a complex language mediated by chemical signals, mostly pheromones. Those can be highly volatile, with low molecular weights and fast diffusion in air or less volatile, heavier, adsorbed on the surface of the body and passed through direct body contact or during food transfer [1]. Honeybee pheromone research has been focused mostly in the compounds present in solvent-derived extracts of glands or body parts, rather than the volatiles emitted in vivo. Although many chemicals have been identified, the understanding of the entire profile emitted by honeybees is crucial to disclose the complex chemical language observed in social hymenoptera.
The aim of this work was to characterize the queen and alarm pheromone of *Apis mellifera iberiensis*, the native honeybee subspecies of Iberian Peninsula, through glandular extraction vs in vivo collection of the emitted volatiles. For queen pheromone, the glandular experiments were conducted in unmated/established mated queens by extraction procedures [2], while emitted volatiles, were collected in vivo by SPME. Similarly, the alarm pheromone produced in the glands was studied by removal and solvent extraction of forager bees sting apparatus, while in vivo experiments were conducted through SPME (with electrical stimulus). Chemical analysis was performed by GC-MS. Concerning the queen pheromone, the results obtained through glandular extraction showed a different composition, with a typical queen mandibular pheromone profile, mainly composed by (E)-9-oxodec-2-enoic acid, (E)-9-hydroxydec-2-enoic acid, methyl p-hydroxybenzoate and 4-hydroxy-3-methoxyphenylethanol, comparing to the volatiles collected in vivo, where the main compounds were octanal, octanol, nonanal, decanal and decanol. On the other hand, the analysis of the sting apparatus extract vs in vivo volatiles showed a more similar profile, with isopentyl acetate, octanol, 2-nonanol, benzyl acetate and n-octyl acetate in its composition.
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[1] Slessor K.N., Winston M.L., Le Conte Y. Journal of Chemical Ecology, 2005, 31(11), 2731-2745.
[2] Slessor K.N., Kaminsky L.A., King G.G.S., Winston M.L. Journal of Chemical Ecology, 1990, 16(3), 4486-4491.

Alarm pheromones mediate plant, pollinator and predator interaction

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Bee alarm pheromones were firstly identified from worker sting gland, which attract other bees to locate intruders and cause the other bees to defense. With the chemical analysis technology developing, more and more alarm components were identified, and they were also proved to have pleiotropic effects not only showing avoidance to risky food resources for mates, but also recruiting mates to repel

predators. These alarm pheromones exist in both all bee (Apis) species and some plants. Whether each component concentration and the persistent of different components combinations encoding different risky levels, how these risk coding system effect on plant-pollinator and predator tri-trophic interactions was still unknow. In this study, we first analysis the alarm pheromones of *Apis cerana*, *Apis dorsata* and *Apis florea* with GC-MS, then we analysis how alarm pheromones effect on intraspecific and interspecific bee species. Will predator, Vespa velutina, eavesdrop on bee alarm pheromones were studied as well. The results turned out that different bee species share similar components in alarm pheromones, while some components are species-specific. Bees could use the bee alarm to alert their mates (intra-specific) but also the other pollinators (inter-specific). Even the predator hornet would eavesdrop on the bee alarm components to hunt pollinators on their food foraging trips. This study may give us a better understanding of bee alarm pheromones mediate the plant-pollinator and predator interaction.

Assessing the attractiveness of hive volatiles to small hive beetles

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Small hive beetle (SHB) is an invasive pest of honeybee.Small hive beetle known to be attractive to volatiles from hive products with the yeast Kodamaea ohmeri and worker bees. Previously we reported ethyl linolenate and ethyl palmitate from fermented pollen dough; oleamide and tetracosane in fermented honey and oleamide and 5-methyl-2-phenyl-1H-indole in workers bees (*Apis mellifera*) as major volatiles. In this study we investigated the attractiveness of the aforementioned volatile organic compounds to SHB by Y-tube olfactometric bioassay. Among these chemicals, ethyl linolenate showed strong attractiveness for male and female SHB. Ethyl palmitate was only attractive at higher concentration and not at lower concentration (0.01-01 mg/ml) for both males and females. On other hand, tetracosane, 5-methyl-2-phenyl-1H-indole and oleamide exhibited repulsiveness to both sex. Ethyl linolenate and ethyl palmitate are also components brood pheromones of honey bees which SHB may utilize these as cue of honey bee hives. Further study might be needed for application of these chemicals as an alternative control method for SHB

BEE BIOLOGY

11 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00
GUT MICROBIOME ROOM 517B

[Lead-off] The honey bee microbiome in health and disease

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The honey bee gut microbiota is integral to host physiology, serving a critical role in host signaling and protection. It is intimately associated with host metabolism and longevity, and functions in host disease prevention, parasite susceptibility, detoxification, immunity and oxidative balance. Constituent or 'core' gut microbiota is hypothesized to mediate susceptibility to disease and ultimately even shape the ecology and evolution of disease states. The stability of the worker microbiota suggests a healthy ecosystem, but like all well-structured communities, the microbiota is beset with cryptic cheaters and opportunists. A variety of biotic and abiotic factors can alter gut microbial balance, leading to decreased host function, decreased longevity, and disease progression. Environmental perturbation or natural variation in the core gut or hive microbiota may contribute to, or even represent the point source for many opportunistic and cryptic disease states associated with honey bee colony decline. Further insight into microbiota structure and function exploring host gene expression, queen and worker aging, floral contributions, larval development, and disease prevalence will further define the critical role of microbiota in honey bee health.

Honey bee gut microbiota regulate the expression of host cytochrome P450 enzymes and contribute to host detoxification ability

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Gut microbiota is known to affect host physiology and a growing number of researches have demonstrated a close relation between insect gut microbiota and insecticide resistance. However, the contribution of honey bee gut microbiota to host detoxication ability has yet to be investigated, especially the effect to host endogenous detoxification pathways. In order to address this question, we compared the expression of CYP6AS1, CYP6AS3, CYP6AS4, CYP6AS10, CYP9Q1, CYP9Q2 and CYP9Q3 in midgut and hindgut between gut microbiota deficient (GD) workers and conventional gut community (CV) workers at mRNA level. The mortality rates of GD and CV workers treated with sublethal dosage thiacloprid or fluvalinate were also measured. Our results show that gut microbiota could promotes the expression of CYP6AS1, CYP6AS3, CYP6AS4, CYP6AS10, CYP9Q2 and CYP9Q3 in midgut, and the survival rate of GD workers are significantly lower than CV workers when treated with pesticide. In addition, we investigated the impact of gut dysbiosis caused by antibiotic treatment on host detoxification ability. Comparisons between antibiotic treated workers and normal workers indicated that gut dysbiosis leads to attenuated expression of cytochrome P450 enzymes (P450s) and reduced survival rate. Taken together, our findings demonstrate a direct relationship between the presence of symbiotic bacteria and the expression of P450s, suggest that honey bee gut symbiont could contribute to bee health through modification of host xenobiotics detoxification pathways and reveal a potential negative impact of antibiotics to honey bee health.

The BeeBiome Data Portal: a web-based resource to facilitate bee microbiome studies by making data findable, accessible and reusable

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Bees are home to a diverse set of microorganisms ranging from protozoa to bacteria to viruses. They can collectively be referred to as the bee microbiome, include beneficial, commensal, and pathogenic species, and play important roles for bee health and disease. With the recent advent of sequencing technologies, scientists have been able to reveal the outstanding genetic and functional diversity of microbial life associated with bees. While these datasets are publicly available from different sources, they have never been catalogued or made accessible from a single resource. Most of these datasets are deposited in different formats and are associated with different meta-data, making it difficult to systematically search for sequence resources and hindering cross-study analysis that would push the field forward. We believe that the scientific community would profit from a web-based platform to share publicly available datasets associated with bee microbiome research. With the financial support of the Eva Crane Trust, we will build the web-based BeeBiome Data Portal. The main aims of this resource will be to (i) catalogue all open-access bee microbiome datasets (mainly sequence data), (ii) enable the mining and integration of these datasets for cross-study analyses, (iii) provide common formats and procedures to deposit novel datasets in repositories like for example the European Nucleotide Archive ENA, and (iv) propose standard methods and terminology. This portal will facilitate collaborative efforts with the ultimate goal to understand the impact of associated microbes and viruses on bees, including

managed species as well as wild pollinators. Moreover, the BeeBiome Data Portal will serve as a mean to reach out to a diverse audience, including beekeepers, the general public, policy-makers and the private sector to inform about bees and their microbes. Informations on the BeeBiome consortium can be found on: https://wp.unil.ch/beebiome/consortium-members/

Impact of nutrition on honey bee immunity, gut microbiota and Nosema ceranae infection

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Apis mellifera colony loss episodes have been reported losses worldwide and have been associated with parasites and pathogens, pesticides and nutritional stress. Nutritional problems are associated to the increase in monoculture areas and the reduction of pollen availability and/or diversity for bees, among others. These conditions may favor the infection by different pathogens, as the microsporidia Nosema spp. In this study, we examined whether the diet influences bee immunity, gut microbiota and the development of Nosema spp., under laboratory conditions. Newly emerged bees were caged and fed ad libitum with two different diets: Eucalyptus grandis (monofloral bee bread) or a mix of at least 18 botanic families (polyfloral bee bread). The experiment was carried out by triplicate and two independent trials were performed. Bees fed on monofloral and polyfloral diets survived almost 60 days under laboratory conditions. No differences between groups were observed. However, at ten days of feeding, E. grandis bee bread decreased the expression level of vitellogenin and genes involved in social and individual immunity (glucose oxidase, hymenoptaecin, lisozym) compared to polyfloral one. The diet also influenced the abundance of some members of the gut microbiota, as bees fed on E. grandis bee bread showed a lower abundance of Lactid Acid Bacteria (Lactobacillus spp. or Bifidobacterium spp.) and higher abundance of Bartonella apis, compared to bees feed on polyfloral diet. In the second trial, bees subjected to different diets were infected with Nosema ceranae spores, and we found that E. grandis pollen favored the multiplication of the microsporidia. These results suggest that nutrition influences social and individual immunity, the abundance of lactic acid bacteria and the development of Nosema spp., contributing to the understanding of the influence of agriculture intensification on colony health.

Engineered symbionts immunize honey bees against colony collapse pathogens

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Honey bees support global agriculture, but also suffer numerous health threats and high hive mortality each year. Pathogens, climate change, and hive management practices all likely contribute to these hive deaths. Deformed wing virus (DWV) and parasitic Varroa destructor mites interact to be among the most significant factors causing these hive losses, and they remain difficult to treat. Here I describe a new approach: engineered symbiotic bacteria from the bee gut. The bee gut microbiome is a simple community of bacteria that plays an important role in host health and nutrition. In this work we have engineered the natural bee gut bacteria Snodgrassella alvi as a platform to express compounds that support bee health while S. alvi lives inside bees. S. alvi producing double-stranded RNA in situ persistently activate RNA-interference, an important component of the honey bee antiviral immune response. We demonstrate these engineered bacteria can be used to control bee gene expression and behavior. Further, we show these engineered symbionts can lower replication of deformed wing virus, thereby improving bee survival, and even kill Varroa mites parasitizing bees. I will discuss how this work will support reverse genetic studies in honey bees, and may one day be used to support the health of bees used in agriculture.

Clothianidin induces microbial gut dysbiosis to honey bees, *Apis mellifera*

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Honey bees are facing a wealth of synergistically interacting stress factors affecting their lifespan, health, and productivity. Clothianidin act on the central nervous system of insects, specifically targeting the nicotinic acetylcholine receptors inducing behavioral, memory and immunity alterations. It is now well documented that functions associated with immune response and behavior are controlled by the intestinal microbiota in insects. Given that clothianidin is persistent in the environment, there is an urgent need to develop alternative and sustainable strategies to mitigate its toxic effects on bee health. The first objective was to identify the host microbiota functional interactions impacted by clothianidin. In vivo studies have been conducted to measure impact of clothianidin exposure on bee survival, behavior, syrup consumption and eubiosis/dysbiosis condition of the gut microbiota. Three concentrations (0.1; 1 and 10 ppb) have been tested. Strikingly, the lowest concentration (0.1 ppb) exerted the most negative impact on bees, showing the highest mortality rate compared to 1 and 10 ppb experimental groups. Moreover, phenotypes changes were recorded in all exposed groups and differences in syrup consumption were

observed across experimental groups. Then, transcriptomic analyses were performed to identify which bee midgut microbiota strains were impacted in terms of functional activity by sublethal concentration of clothianidin. The second objective was to select endogenous probiotic candidates being able to degrade clothianidin into innocuous metabolites. Endogenous microbial communities from host gut microbiota were observed to improve host's resistance against bacterial pathogens and intracellular parasites, both in mammals and insect models. Our pesticide quantification reported a complete degradation of clothianidin in contact with some probiotic candidates. The third objective was to measure under in vivo conditions effect of select endogenous probiotic candidates administrated to bees exposed to clothianidin to test if the selected strains will help to restore the impaired functions. Our results highlighted promising endogenous bacterial probiotic candidates to develop a probiotic formulation mitigating the negative impact of neonicotinoid exposure on bee colonies. More specifically, administration of one of our probiotics significantly improved the survival rate of bees exposed to clothianidin compared to control bees (i.e. supplemented with sugar (1:1)).

Sensitivity of honeybee gut microorganisms to antimicrobial substances present in beekeeping

V. Strogolov
Strong Microbials, Inc, Milwaukee, USA

The honey bee beneficial microbiota represents a key variable in honey bee colony health. Antimicrobial and chemical treatments wipe out microbial communities and can cause dysbiosis in bees. Lactic acid bacteria, including Lactobacillus, are most abundant in honey bee hind gut where they mitigate toxin exposure, nutrient absorption, and stimulate innate immune responses. Synergistic or antagonistic interactions between microbial and antimicrobial factors present in beekeeping warrants greater exploration. Sensitivity of honeybee gut lactic acid bacteria to antimicrobial substances in bee environment: antibiotics tetracycline, tylosin, lincomycin, fumagillin, pesticides glyphosate and fipronil, and plant extracts, is addressed in this work.

SYMPOSIUM 13:00-15:00
LEARNING, COGNITION AND BEHAVIOUR IN STINGLESS AND HONEY BEES ROOM 517B

[Lead-off] Recent advances in the neuroethology of olfactory perception and learning in honey bees

J.C. Sandoz
EGCE, CNRS, University Paris-Saclay, Gif-sur-Yvette, FRANCE

For many years, honeybees have been an influential model in the study of olfactory perception and learning in insects. Recently, we made remarkable progress in our understanding of how honeybees perceive, learn and use olfactory cues in their mating and social behaviors. First, I will address the use of olfactory cues by honeybee drones during mating and show that both drones and virgin queens are attracted to volatiles emitted by mature drones in olfactory attraction tests. I will discuss the possible implications of these findings for the formation of drone congregations and honeybee mating, and will detail our current understanding of how such cues are processed in the drone brain. Second, I will present our recent discovery of a new social learning form in honeybees. Our data show that a simple social contact with a nestmate can be perceived as a rewarding stimulus by other bees, so that an initially neutral odorant, when associated with such social contact, triggers an appetitive response (proboscis extension). Our results suggest that this social reinforcement is mediated by bees' intricate antennal communication. I will discuss how this process may facilitate resource exploitation by bees.

[Lead-off] Keeping The Stingless Bees

D. Roubik
Smithsonian Institution, Tropical Research Institute, Balboa, PANAMA

The stingless honey bees have something for everyone, thus keepers of Stingless Bee colonies find themselves being pulled in different directions. However, never have I known a Stingless Bee keeper to return bee colonies to the forest, carrying coal to Newcastle.
Research with meliponines, with over 600 named species to choose from, has a bright future. Stingless honey bees live in most the tropical and some of the temperate world, and their range is expanding, both with climate change and humans. What do they have? And why are they receiving attention now? For one, if you gave up on AHBs in the Americas, they offer an alternative. You may feel you are participating in conservation, when you remove a nest from a tree that is about to be falled and burned. Meliponines may validate your cultural history, in the tropics, as a traditional source of alcoholic beverage, food, sweetener or medicine. Meliponini are a low-cost positive input for any family, community or townsite, because they offer harvestable products, pollinate or at least visit plants of all kinds, do not require much maintenance, and can be propagated. They are neither livestock nor to be treated similarly to honeybees— both facts that new beekeepers are often slow to learn.
The first parasite, a bacterium, of Stingless Bees was recently detected, in Australia. One wonders what further developments may come from altering the number, density or genetic makeup of such bee populations. Although beekeepers have made meliponines highly profitable as crop pollinators, and scientific data confirm medicinal utility (as with the manuka honeys harvested by exotic honeybees), the lawyers of land use and regulations have also discovered them. Legislation makes it increasingly difficult to use, transport, buy, sell,

harvest or even study the Stingless Bees. How soon will a reasonable scheme evolve?
Stingless Bee keepers must be innovators, scientists, diplomats, agronomists and economists. There is legislation, competition, basic research, commercial production pressure, supply and demand, health concerns and finally, sustainability. Sustainability is only relative. Does anyone know whether Stingless Bees will survive in the world 100 years from now? We shall see.

Collective Foraging Behavior is Driven by Individual Variation in Learning, Waggle Dancing in Honey Bees

C. Cook [1], N. Lemanski [2], T. Mosqueiro [2], J. Gadau [3], C. Ozturk [1], B. Smith [1], N. Pinter-Wollman [2]
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In the honey bee society, the foragers must collect enough food to feed the whole colony. For efficiency, foraging honey bees divide this task into explorers, who search the landscape, and exploiters, who revisit the same location. These individual differences are driven in part by how these bees learn. Explorers learn to ignore familiar odors, exhibiting high latent inhibition (LI). Exploiters learn both novel & familiar odors, exhibiting low LI. LI behavior is genetically heritable, allowing us to select for high & low LI in queens and drones, generating workers of known LI. Using colonies consisting of different proportions of workers with only high LI, only low LI, & a 50/50 mix, we found that these colonies differed greatly in their collective foraging behavior: Visitation increased after high LI bees discovered a feeder. Low LI bees foraged at both novel and familiar feeders while high LI bees surprisingly preferred the familiar feeder. Differences in waggle dance performance drives foraging behavior: High LI dancers performed more intense dances than low LI dancers. Our work shows that individual genetic variation can lead to major ecological consequences for societies.

Uncovering nurse task cycles using high resolution behavioral data

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Nursing behavior has been extensively studied in social insects, particularly in honey bees. This is in part because of the important role nurses have in rearing new nestmate workers, drones, and queens which is essential to colony growth and reproduction. Our knowledge of nurse behavior outside of feeding and caring for brood however, is much less known. In this talk, we present an overview of nurse task profiles, interaction rates, and behavioral consistency using high resolution behavioral data (25h over 3.5 days of second-by-second behavior) for 12 nurse aged workers. Using these data, we explore the hypothesis that, because caring for brood expends nutritional reserves, which must be replenished by consuming stored honey and pollen and, once fed, nurses need time to rest and metabolize consumed food to produce royal jelly, nurses will perform task cycles sequentially rotating from brood care to feeding to rest, and back to brood care. Our results of short- and long-term patterns of task sequences show cycles alternating between nursing and resting, with idiosyncratic bouts of feeding interspersed throughout. Nurses showed consistent individual behavior and significant differences among nurses, including in feeding bout frequency and duration, which may be regulated by interaction rate with nestmates and the queen. Ultimately, our results provide insight into the role of nurse behavior in using colony inputs (collected and stored resources) to support developing brood.

Search of Bacillus spp. in *Nannotrigona testaceicornis* (Lepeletier, 1836)

L. Gavazzoni [1], D. Galhardo [1], M. Felgueira Pavanelli [2], V.A. Arnaut De Toledo [1]
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The stingless bees also known as meliponíneos, have a greater diversity of species, being considered fundamental for the diversity of vegetal species. In order for the hive to be healthy and have good productivity, the bees make mutualism / symbiosis interactions with specific microorganisms. One of the most commonly found microorganisms is bacteria of the genus Bacillus, which was cited in several studies among species of stingless bees associated with workers and queens. This study aimed to identify these microorganisms in the stingless bee *Nannotrigona testaceicornis* (Iraí or Black Jataí), known for its potential pollinator, mainly in greenhouses. Twenty bees were collected from each of the six colonies of N. testaceicornis. The samples were collected manually aseptically, using gloves and sterile collection bags. The samples were washed with sodium hypochlorite and sterile water and then macerated and the whole contents were homogenized in tubes containing Brain Heart Infusion (BHI) broth, obtaining a 10% solution, which was incubated in an oven at 37 ° C for six hours. Pre-enriched samples were inoculated in triplicate on plates containing BHI agar, Trypticase Soy Agar (TSA), Nutrient Agar (AN), MacConkey (MC) agar and Mannitol Salt Agar (MAN), incubated at 37 ° C for 24 / 48h. Different colonies were submitted to Gram staining (1884) and biochemical tests of catalase, oxidase, motility and 6.5% NaCl. The isolated samples presented catalase positive, sporulated bacilli morphology and Gram positive staining, with variations in sizes and arrangements, indicating the presence of different species or lineages previously identified as Bacillus spp. Species of this genus are known to have antimicrobial potential as well as aid in the pre-digestion of pollen. The results obtained confirm the symbiosis relationship between the sporulated Gram positive bacteria and the stingless bee in study, since they can help the colony health avoiding contamination by pathogens. In addition, this study opens new means of research regarding the production of probiotics for improvement in the production and development of this and other species of bees.

Sticking together: Resin use and social immunity in honey bees and stingless bees

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Social insects employ behavioral, physiological, and organizational strategies – called social immunity – to defend against pathogens within the crowded colony nest environment. One form of social immunity is the collection of antimicrobial resins. Honey bees and stingless bees use resin, or "propolis" to seal the cracks and crevices found in the hollow tree cavities where they nest. Propolis-rich nest environments have been shown to reduce clinical signs of honey bee brood diseases and modulate the immune response of individual bees, allowing otherwise chronically activated immune systems to rest and respond more effectively to acute threats. Propolis use in honey bees has been researched extensively, and the therapeutic value of propolis to humans is well-established, but comparatively little is known about the benefits of propolis to stingless bee health. Tree-nesting stingless bees construct propolis-rich structures (e.g. batumen) in the hollow tree cavities where they naturally nest. However, some stingless bee management practices may reduce propolis deposition. If propolis-rich environments support stingless bee immune systems (as occurs in honey bees), then decreasing propolis deposition will likely have negative consequences for stingless bee health. I review our current knowledge of propolis use in honey bees and discuss opportunities for applying honey bee research methods to the stingless bee system to address unanswered questions around resin use and social immunity in stingless bees.

SYMPOSIUM — 15:30-17:30
WINTER SURVIVAL IN COLD TEMPERATE & TROPICAL CLIMATES — ROOM 517B

[Lead-off] Wintering Bees Indoors and Outdoors in Canada: Implications and Opportunities for Managment of Parasites and Pathogens

R. Currie
Dept. of Entomology, University of Manitoba, Winnipeg, CANADA

In Canada honey bees are often wintered in regions where climate extremes during winter are beyond the normal range of the European Honey bees making successful wintering a challenge. Canadian beekeepers have developed systems for wintering bees both outdoors under extremely long, cold winters, and within climate controlled buildings that provide constant climatic conditions for wintering bees for over 5 months of the year. Winter is a period of high stress and thus interactions with the myriad of other stressors affecting bees often result in much higher levels of winter loss than would be expected. Interactions among pests and pathogens of bee colonies differ depending upon the wintering environment (Indoor or outdoor). Indoor wintering offers many advantages in mitigating the impacts of multiple stressors that affect colony survival. Flexibility in the control of temperature, humidity, CO2, ventilation and the ability to feed and treat colonies in mid-winter can result in significantly better winter survival when colonies are wintered indoors relative to those with similar pre-winter pathogen profiles that are wintered outdoors. This presentation will address recent advances in our knowledge of how differences in the wintering climate can affect the interactions among parasites and pathogens and what management steps can be used to mitigate pest and parasite populations during winter storage.

The effects of probiotics on winter survival, spring development and gut microbiota of the honey bee (*Apis mellifera*) in Quebec, Canada

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The honey bee harbours a simple yet essential microbiota that contributes to digestion, immunity and protection of the gut lining. Climate, diseases and antibiotics are known to disrupt the balanced microbiota and cause dysbiosis, which affects honey bee health. The use of probiotics is now considered by many as a sustainable strategy to improve colony health. In fact, studies have shown that some probiotic strains could not only prevent dysbiosis but also enhance brood development, honey yield and pathogen tolerance. However, none of them investigated the benefits of probiotic supplementation under a cold climate, where colony winter survival and spring buildup are two critical moments for beekeepers. To this purpose, between October 2017 and May 2018, forty-five honey bee colonies were given sugar syrup supplemented with a commercial probiotic strain (Pediococcus acidilactici MA 18/5M or Saccharomyces cerevisiae boulardii) or an endogenous bacterium (Parasaccharibacter apium). A fourth group was treated with the antibiotic Fumagillin-B® in order to compare this medication with the probiotics on Nosema sp. spore load. Colony performance was assessed using brood coverage, number of frames covered in bees and hive weight. The Varroa destructor population was estimated with daily drop on sticky boards at key moments during the experiment. A molecular analysis of the microbiota determined the impacts of the treatments on the bacterial community inhabiting the midgut, including the microorganisms causing European and American foulbrood. The results of this project will provide us with useful information to develop a safe and effective probiotic formulation specific to the Canadian beekeeping industry.

Investigating the Regulation of Hypopharyngeal Gland Activity in Honeybees Under Overwintering Conditions via Morphologic Analysis Combined With iTRAQ-Based Comparative Proteomics

K. Wang, T. Ji
Yangzhou University, Jiangsu, CHINA

The management of overwintering honeybee colonies is pivotal for the apiculture industry in China. To investigate the molecular mechanisms involved in the seasonal regulation of the development of *Apis mellifera carnicas* hypopharyngeal gland (HG), transmission electron microscopy (TEM), and large-scale proteomics (isobaric tag for relative and absolute quantification; iTRAQ) were employed. In total, 3,025 proteins were identified at two-time points: winter (constituting group A) and early spring (constituting group B). Among them, 24 proteins were quantified and compared to identify their differential expression patterns. The TEM profiles and iTRAQ analysis results might aid in understanding the molecular mechanisms associated with the seasonal changes in honeybee HG. For example, the acinar cells of spring worker honeybees had more mature organelles. Furthermore, the ribosomal structures and protein biosynthesis were strengthened. However, the amount of exocrine royal jelly produced could not have satisfied the demand of the feeding larvae and queen. Honeybees might suffer reduced immune function during their feeding stage. The present study elucidates the regulatory mechanisms of HG on overwintering and spring arousal in honeybees. The results might provide a practical and theoretical guideline for the overwintering management program of the apicultural industry.

Aspects and representativeness of monitoring colony losses in the Czech Republic

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The Czech Republic is a middle European country with a population of about 11 millions citizens. Beekeeping is a popular hobby there. There are approx. 60,000 registered beekeepers with more than 650,000 colonies. Detailed and area-wide monitoring of honey bee colony losses started in spring 2014 when the pilot survey based on the COLOSS standardised questionnaire was first performed. Since then the standardised COLOSS questionnaire is distributed to beekeepers every springtime. Different approaches are being used for dissemination, e.g. internet, beekeeping journals, meetings with local beekeeping clubs etc. The number of participating beekeepers slightly and continuously increased since the beginning and the response rate of the survey fluctuates around 2 % of the total number of beekeepers. However, the spatial distribution of responses varies each year of the survey.
We can clearly detect clusters of respondents in particular areas, whereas in some areas responses are entirely missing. This unequal coverage together with a small number of respondents in some parts causes problems in data evaluation and interpretation of the results. On the other hand, we were provided with the anonymised official database of beekeepers, apiaries and colonies from Czech authorities. This mandatory registration of beekeepers and apiaries including the number of wintered colonies can significantly help in data processing and trustworthiness of results. Comparison of authority data of wintering colonies and a number of beekeepers with data acquired by questionnaires can help us to define the representativeness of answers and finally define the representativeness level for publication of results. In this contribution, we would like to present the results of a five-year study including geoinformatically processed data in the form of maps. The attention will be put in the level of representativeness and include uncertainty in results.

Mass storage of honey bee queens overwintered at different temperatures in Canada

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There is a high demand for quality queens to replace dead colonies or deficient queens after wintering in Canada. Replacement queens are imported mainly from US queen breeders but there is growing concern amongst Canadian beekeepers regarding the sustainability of the industry and the importance to increase self-sufficiency. The goal of this project is to successfully overwinter large number of mated queens from September to April. Our research project tested different methods to store large numbers of individually caged mated queens within a banking mother colony. Our project aims to assess the potential of queen overwintering systems at temperatures below and above cluster formation to maximize queen survival and quality. This project will test 4 queen storing/banking methods from September to April. A total of 600 young mated queens produced during the last breeding cycle in August have been wintered in environmentally controlled rooms at the following conditions: Control Group 1) Queens stored individually within colonies at 4-6°C; experimental Group 2) Queens stored individually in small cages and grouped together within a queenless colony (bank) below cluster formation temperature at 4-6°C; experimental Group 3) Queens stored individually in small cages and grouped together (queen banks) within a queenless colony near cluster formation temperature at 10 -13°C; experimental Group 4) Queens stored individually in small cages and grouped together within a queenless colony (bank) above cluster formation temperature at 15-18°C. From September 2018 to April 2019, survival of queens and various quantitative physical characteristics of queen quality are assessed: weight, size and sperm viability. A subsample of surviving wintered queens are introduced in colonies to evaluate their performance during the following season: colony development and honey production.

4-compartment hive with cruciform partitions (Palamarchuk hive) for excellent nucs wintering and their intensive spring development with the aim of early nucs production

O. Komisar
Union of Beekeepers of Ukraine, Kyiv, UKRAINE

Ukraine is the country with relatively long winter (150 days without cleansing flights) and winter losses of bee colonies usually are more than 10%.
According to Standards of early nucs for sale before the 10-th of May, nuc must have 3 Dadant (or 4 Langstroth) frames of brood and one honey frame.
It was elaborated the design of hive, where 4 nucs formed the common winter cluster near the center of crusiform partition. They overwintered excellently and had better spring development, than in usual colonies, in majority cases. The clear explanation of this phenomenon is not yet developed.
Honeybees of every nuc have rigid binding to the center of the common winter cluster, they can't shift to another honey frames and cluster is able to move only upwards. Therefore, the frames must be high, but, from the other hand, they ought to be standard, as in spring we will sell them.
It was the Ukrainian beekeeper Valery Palamachuk, who, for the first time, proposed to use these hives for early nucs production and to apply rotated to 90° Langthroth frame in them. And this 4-compartment hive with cruciform partitions was called after him.
There are two strategies for use of such hives: the main, propoused by Palamarchuk, — production of early nucs, and the second — strengthening of other colonies before the main honey flow.
Several modifications of Palamarchuk hive with 6, 8, 10 or even 12 frames in each hive compartment were tested. This hive was also produced of different materials: 10 mm plywood, OSB , 20 or 30 mm styrofoam and usual 30 mm wood.
All beekeepers, who started to exploit this hive, obtained rapid growth of quantity of colonies at their apiaries and became the sellers of early nucs. Several cases of extremely low winter honey consumption of nucs in Palamurchuk hives were observed and this phenomenon needs special scientific investigation.
Author hopes that this type of hives will be useful for beekeepers in cold Canada.

Thermopreferendum of the honey bees and high temperature wintering

O. Komisar
Union of Beekeepers of Ukraine, Kyiv, UKRAINE

High-temperature wintering of bees (abbreviated HTW) means wintering at unusually high temperature (15-25°C) with the obligatory cold zone and water supply. The simplest realization of method is the hight observation hive in a heated room with the tunnel for bees to go outside. The "ABC and XYZ of Bee Culture" (1959) wrote "bees in observational hives badly winter in warm rooms, since they cannot form winter cluster". In fact bees can winter without formation of the winter cluster, but they die because of dehydration. Providing bees with water in gravitational drinkers normalizes wintering. There are two ways of HTW: either to keep the bees in a warm room with the possibility of their exit outside through the cold tunnel, or to keep them outside and locally heat by electricity upper part of the hive with the cold zone below.
Thermopreferendum is the reaction of organisms moving to the zone of preferred temperatures, where their energy losses are minimal.
We studied the thermopreferendum of bees in winter in the gradient device with a vertical temperature gradient. Bees located in temperature range of 15-35°, and queens located in the area of 25-35° only. We observed short-term moving outside of these zones, but they didn't stop there. In the winter cluster, part of bees overwinter in non-optimal conditions and therefore their energy losses (food consumption) are not minimal. It is possible to reduce the total food consumption using the HTW.
My book "HTW of honey bees" (168 pages, in Russian) was published in Kiev in 1994, but only few beekeepers tested HTW. The results were impressive. Beekeeping should have a backup system of wintering bees in case of their weakening from any reasons and this method is HTW. There is an urgent need to bring the HTW to a simple implementation and popularize it.
Beekeeping should have a backup system of wintering bees in case of their weakening from any reasons and this method is HTW. There is an urgent need to bring the HTW to a simple implementation and popularize this method of nucs wintering and even baby nucs with reserve queens.

BEE HEALTH

09 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

IMPACT OF PESTICIDES ON BEES I ROOM 517C

[Lead-off] Honey bee reproductive toxicology: possible effects of neonicotinoids on workers, drones, and queens

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[1] *Auburn University, Auburn, USA,* [2] *University of Bern, Bern, SWITZERLAND*

Sub-lethal effects of neonicotinoid pesticides on honey bee lifespan, behavior and physiology are relatively well-documented, unlike their possible impacts specifically on reproduction. Here we explore royal jelly, worker hypopharyngeal glands, drone sperm quality, and queen sperm storage, among other endpoints, to better understand how neonicotinoids can potentially affect honey bee reproduction. To do this, we employed a common experimental treatment regime that consisted of exposing 20-30 honey bee colonies to pollen patties for two brood cycles (42 days) during spring; half the colonies received pollen patties that were spiked with environmentally-relevant concentrations of thiamethoxam (4.5 ppb) and clothianidin (1.5 ppb) in order to create two colony-level treatment groups: 1. Neonicotinoids and 2. Controls. Multiple individual honey bees, as well as queen cells, were reared in each experimental colony to allow for comparison between the two treatments. Over the past five years, our studies have revealed that neonicotinoids can negatively influence a variety of honey bee characteristics related to reproduction, both in the laboratory and the field. This work highlights the importance of evaluating honey bee queens and drones, in addition to workers, during agro-chemical risk assessments and basic toxicology studies.

Grooming behaviour in honey bees is compromised by the interaction between sublethal exposure to neonicotinoids and Varroa destructor

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[1] *University of Guelph, Guelph, CANADA,* [2] *Purdue University, West Lafayette, USA*

Honey bees (*Apis mellifera*) are exposed to a number of stressors, but little is known about the effect of their interaction on behaviours associated with defense mechanisms. This study evaluated the interaction between sublethal doses of a neurotoxin, clothianidin, and the ectoparasite Varroa destructor by assessing grooming behaviour, the number of differentially expressed genes (DEGs) and DWV levels in the brains of bees performing grooming behaviour. An increase of DWV levels was observed in bees treated with the lowest dose of clothianidin, and in bees parasitized by V. destructor. The highest dose of clothianidin showed the highest number of up-regulated DEGs and an effect on biological pathways related with neurodegenerative disorders, like Alzheimer's, which could explain disruption in neural processes affecting the ability of the bees to perform intense grooming. Also, an interaction between clothianidin and V. destructor reduced the proportion of bees performing intense grooming behaviour and a reduced the number of up and down-regulated DEGs. The study revealed a complex interaction between clothianidin and V. destructor that resulted in non-additive effects.

Differences in larval pesticide toxicity across honey bee (*Apis mellifera*) stocks

J. Milone, D. Tarpy
North Carolina State University, Raleigh, USA

Honey bee (*Apis mellifera*) colonies retain residues from many chemicals which results in exposure to multiple pesticides during sensitive developmental stages. The harmful effects of these exposures depends on the susceptibility of bees to a given dose. We compared pesticide susceptibility among 8 North American honey bee breeding stocks using a larval oral exposure assay. To test field-realistic exposures, we selected a mixture of 7 commonly detected insecticides, fungicides, and an herbicide using previously reported pesticide residue data from commercial colonies. We chronically administered diet spiked with the treatment mixture at 4 dose levels during worker larval development in vitro. A Hazard Quotient (HQ) was used to quantify toxicity from the multi-pesticide mixture at each concentration and survivorship to the exposure was compared among the offspring from 30 colonies. The median lethal HQ values for each stock showed a gradient with three clusters of susceptibility. Larvae from stocks know for artificial selection were among the most susceptible to pesticide exposure, while larvae from distinct and unmanaged stocks were found to have higher tolerances. These findings highlight the need to account for stock differences when testing and reporting honey bee toxicology measures. Future work is needed to elucidate the mechanisms driving differential susceptibility to pesticides in honey bees.

Chronic Exposure to Neonicotinoids and Honey Bee Health

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Experiments linking neonicotinoids and declining bee health have been criticized for not simulating realistic exposure. Here we quantified

the duration and magnitude of neonicotinoid exposure in Canada's corn-growing regions and used these data to design realistic experiments to investigate the effect of such insecticides on honey bees. Colonies near corn were naturally exposed to neonicotinoids for up to 4 months - the majority of the honey bee's active season. Realistic experiments showed that neonicotinoids increased worker mortality and were associated with declines in social immunity and increased queenlessness over time. We also discovered that the acute toxicity of neonicotinoids to honey bees doubles in the presence of a commonly encountered fungicide. Our work demonstrates that field-realistic exposure to neonicotinoids can reduce honey bee health in corn-growing regions.

Bumblebee (*Bombus terrestris*) colonies exposed to field-realistic doses of imidacloprid produce larger workers and abnormal behavior in-hive

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The neonicotinoid insecticide imidacloprid (IMD) is one of the most applied agrochemicals in the world. As a systemic insecticide, its residues can be found in nectar and pollen of flowers from treated crops and their surroundings, usually at levels around 10 ppb. *Bumblebee*s are likely to be exposed to pesticides when collecting resources from flowers close or within agricultural areas. Chronic exposure to neonicotinoids has significant consequences in *Bumblebee* colonies, including reduced growth and productivity. Bees exposed to IMD show hampered foraging abilities, weakened immune system, and abnormal behavior. The division of labor in *Bumblebee* colonies relates to body size with larger workers typically engage in foraging tasks whereas their smaller sisters typically take in-nest duties such as brood tending. In this study, we investigated the effects of field-realistic doses of IMD on the behavior of *Bumblebee*s, as well as the possible role played by the behavior of exposed queens in the determination of worker bees' body size. We tested 14 colonies that fed on sucrose solution supplemented with 0.0, 1.0, or 10.0 µg/L of IMD. We recorded daily the body size of all adult bees emerging in these colonies for 10 weeks, as well as performed observations of queens' behavior in-hive. At the end of the experiment, control colonies weighed 5.65 and 4.59% higher than colonies treated with 1 and 10µg/L IMD, respectively (Generalized Estimating Equation test – Wald Chi-Square: 11.756, df= 2, P= 0.003). Worker bees emerging in the IMD treated colonies were larger compared with control colonies (WCS: 43.43, P< 0.001). The same effect was observed for males (WCS: 30.960, P< 0.001). The exposure to IMD in hive also produced a significant decrease in the time queens performed brood-care related behavior (WCS: 48.279, P< 0.001). Our findings add to evidence of IMD as a significant disruptor of colony's homeostasis. This is the first report of a xenobiotic producing an increase in the body-size of worker bees and reproductive individuals. Our data also corroborates to previous findings that queen's behavior may play a significant role as a mediator of worker bees' body size in hive.

Effect of sublethal exposure to thiamethoxam and Nosema ceranae infections on the survival and immune response of the stingless bee *Melipona colimana* (Hymenoptera: Meliponini)

J.O. Macias-Macias [1], A. De La Mora-Peña [2], E. Guzman-Novoa [1,3], J.M. Tapia-Gonzalez [1], F. Contreras-Escareño [1], T. Petukhova [1,4], C.A. Medina-Flores [2]

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Several studies have shown that neonicotinoid insecticides and the microsporidium parasite Nosema ceranae, affect the health and lifespan of honey bees (*Apis mellifera*). However, there is no information about the effect of these stressors on other valuable pollinators such as the stingless bees (Hymenoptera: Meliponini). This study was conducted to examine the effect of one neonicotinoid insecticide (thiamethoxam) and Nosema ceranae on infection development, survivorship and cellular immune responses of the stingless bee *Melipona colimana* Ayala. Newly-emerged bees that were maintained in an incubator, were individually subjected to one of the following treatments: sublethal dose of thiamethoxam (0.4 ng/bee) in sugar syrup, N. ceranae spores in sugar syrup (50,000 spores/bee), thiamethoxam and N. ceranae, and control (bees that received sugar syrup only). Mortality was recorded daily and Nosema infection intensity, as well as hemocyte counts, were determined at 14 days post-treatment in the surviving bees. N. ceranae developed infections in 70% of the inoculated bees at levels of 500,000 spores/bee in the group that was treated with spores only. However, in the bees subjected to the thiamethoxam and N. ceranae treatment, infections were below 250,000 spores/bee. N. ceranae infections did not affect survivorship, but thiamethoxam alone or combined with N. ceranae, significantly increased bee mortality. Hemocyte counts were significantly lower in N. ceranae infected bees than in the control, whereas no reduction in hemocyte numbers were detected in bees treated with thiamethoxam alone or in combination with N. ceranae. These results suggest that N. ceranae may infect and multiply in stingless bees, that exposure to sublethal doses of thiamethoxam is toxic to M. colimana and that thiamethoxam inhibits N. ceranae multiplication and its effect on cellular immunity. These findings have implications on pollinators' conservation.

Effects of Chlorothalonil on Honey Bee Cell Health and Function

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The broad-spectrum fungicide, chlorothalonil, is used widely on crops visited by honey bees. Contact with treated floral resources may provide one point of exposure to the fungicide; however, honey bee adults and immatures may also become exposed to residues found in the hive. Chlorothalonil residues are a frequent contaminant of pollen and wax sampled from the hive and have been detected in honey bee tissues (max concentration – 870 ppb). Due to the prevalence of this fungicide in the hive and the environment, we evaluated its potential to affect honey bee health. Our approach was to use in vitro systems composed of primary hemocyte cultures and the continuous cell line, AmE-711. We measured cell respiration and immune gene expression, markers of cell health, in response to fungicide exposure. Our findings suggest that exposure to chlorothalonil negatively affects basal respiration and spare capacity of honey bee cells in culture. Moreover, exposure to the fungicide disrupts the expression of genes associated with the cellular and humoral immune responses. Exposure to chlorothalonil could reduce respiration rates of cells through increased production of reactive oxygen species and disruption of the mitochondrial membrane potential. The mechanisms underlying observed changes in cells are not well understood, and further studies are needed to establish effects on live bees and colonies. Application of our in vitro system composed of honey bee cells will promote further insights into the impacts of chlorothalonil, as well as other toxicants, on honey bee health at the cell level.

SYMPOSIUM — 10:00-12:00
IMPACT OF PESTICIDES ON BEES II — ROOM 517C

Alteration Of Survival And Oxidative Balance Induced By Subchronic Exposure Of Overwintered Honeybees To Insecticide, Fungicide And Herbicide Combinations

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Honeybees are exposed during their foraging activities to a large variety of pollutants including pesticides. Overwintered honeybees are, as a result, subjected to chronic sublethal exposures of pesticide mixtures. To explore the impact of different pesticide combinations on winter honeybee survival, we chronically exposed adult overwintered honeybees during 16 days to low concentrations (0.01, 0.1, 1 and 10 µg/L) of imidacloprid (neonicotinoid insecticide), difenoconazole (triazole fungicide) and glyphosate (amino-phosphonate glycine herbicide) alone or in binary and ternary mixtures. The survival rates and food consumption were recorded daily. In order to measure the impact on oxidative balance, we measured the activity of six enzymes involved in the antioxidative defenses in the honeybee's head, midgut and abdomen and oxidative stress damages by quantifying midgut lipid and protein oxidation accompanied with midgut histological analysis. Mortality rates suggest that the effects on bees are not dose dependent; the concentration of 0.1 µg/L induces the highest mortality rates, followed by the concentrations of 1 and 10 µg/L. In addition, pesticide mixtures have higher impact than pesticides alone. These effects vary according to the combinations and the concentrations: An additive effect was observed with the insecticide-fungicide mixture at the 4 different concentrations and at 1 and 10 µg/L for the ternary mixture, whereas a synergistic effect occurs with the ternary mixture at 0.1 µg/L and the insecticide-herbicide mixture at 0.01, 1 and 10 µg/L. The physiological markers, and the lipid and protein oxidation, reveal a disruption of oxidative balance at 1 µg/L with an increase of the activities of glutathione-S-transferase, glutathione reductase and glutathione peroxidase in the head, and superoxide dismutase, glucose-6-phosphate dehydrogenase and glutathione-S-transferase in the midgut. Conversely, a decrease is observed for superoxide dismutase in the head and catalase in the midgut. These results suggest that these three types of agrochemicals with different mode of actions may have common molecular targets in honeybees, leading, when combined, to greater alterations and drastic honeybee mortality.

Transportation sensitizes honey bees to insecticides

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Honey bees are critical pollinators whose managed populations face global declines due, in part, to an increased use of insecticides. Transportation of honey bees is necessary all over the world to meet the demands of fruit and vegetable pollination. Previous studies suggested transportation can adversely affect honey bee physiology but it is not clear whether transportation can affect honey bee sensitivity to pesticides. We hypothesize that transportation weakens honey bees such that they are more prone to insecticide poisoning. We found that transportation and mock transportation both sensitize honey bees to neonicotinoid insecticides, imidacloprid and thiamethoxam. We then tested whether this is because transportation makes bees age faster and that older bees are more sensitive to insecticides. Indeed, significantly higher juvenile hormone titers were found in mock transported bees than that in control bees, suggesting that transported bees aged faster. In addition, old bees were more sensitive to pesticides than younger bees. These results suggest the mechanisms of sensitization are due to transportation making bees age faster and aged bees being more vulnerable to insecticides. Thus the widely-

practiced transportation of honey bee colonies sensitizes them to insecticides, increasing their risk of pesticide poisoning. Further studies are needed to provide measures to counter these effects.

Thiamethoxam sublethal exposure reduces cellular immunity and restrains Nosema ceranae infections in the honey bee, *Apis mellifera*

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Honey bee (*Apis mellifera*) health can be compromised by single factors such as neonicotinoid insecticides and the microsporidium parasite Nosema ceranae, but there are few studies aimed at analyzing the combined effects of multiple stressors in the health of honey bees. The objective of this study was to examine the effect of a neonicotinoid insecticide (thiamethoxam) and N. ceranae on the development of infection, survival and cellular immune responses of honey bees. We used newly emerged bees kept caged in an incubator that were randomly subjected to either of three treatments. Treatment 1: sublethal dose of thiamethoxam (0.4 ng/bee) in sugar syrup, treatment 2: N. ceranae administered in sugar syrup (50,000 spores/bee), treatment 3: thiamethoxam and N ceranae, and treatment 4: control (bees that only received sugar syrup). Mortality was recorded daily and N. ceranae infection intensity, as well as hemocyte counts, were determined at 18 days post-treatment in the surviving bees. N. ceranae developed infections in 40% of the inoculated bees at levels of ca. 9'000,000 spores/bee in the group that was treated with spores only. However, in the bees subjected to thiamethoxam or the combination of thiamethoxam and N. ceranae, only 18% of the inoculated bees developed infections and the intensity of infections were significantly lower than in Nosema-infected bees (<3'000,000 spores/bee). Neither N. ceranae infections nor thiamethoxam alone or combined with N. ceranae, significantly increased bee mortality. Hemocyte counts were significantly lower in all experimental groups with respect to the control. These results show that N. ceranae and sublethal exposure to thiamethoxam reduce cellular immune responses in *A. mellifera* and that thiamethoxam retrains N. ceranae multiplication in honey bee guts. The implications of these results will be discussed.

Diversification of Floral Resources in Pigeonpea to Reduce the Impact of Insecticides on Honey bees

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Honey bees are the most important pollinators as they play a unique role in maintaining the nativity of a particular ecosystem, including agricultural, horticultural and forest ecosystems. Now there is a frightening decline in the population of honey bees around the world because of the conversion of forest and landscape areas into urban areas and intensive mono-cropping which causes loss of important fundamentals for bees. Another significant reason is over usage of pesticides which lead to the decrease in honey bee pollination around the world. The economical value of pollinators has been estimated to the extent of 153 billion euros in different crops around the world
An experiment was conducted to diversify the floral resources in pigeonpea to enhance the pollination by honey bees and to reduce the toxicity of insecticide under field condition at Vijayapur and Bagalkot districts during 2018-19. Buckwheat (Fagropyrum esculentum) which is grown throughout a large area of Asia and South East Asia can sustain huge number of insect pollinators belonging to four orders. The most abundant pollinators among the hymenopterans are the little bee, *Apis florea*, *A. dorsata* and *A. cerana*. Since, all the three species of honey bees are the common pollinators of both pigeonpea and buckwheat, the later was selected as a marginal crop around pigeonpea to support the pollinators The floral resource provisioning with buckwheat around pigeonpea has greatly benefited pigeonpea interns of immediate restoration of pollinators on pigeonpea after the application of insecticides. The normal bee activity was assumed on the 3-4th day after application of insecticides with marginal crop against 6-7 days in the treatment without the marginal crop. Among the insecticides used for the management of pod borer in pigeonpea, Neem Seed Kernel Extract and chlorantraniliprole have restored maximum bee activity at a faster rate. Further, A.dorsata assumed the crop immediately followed by *A. cerana* and A. florea. Yield and yield attributing characters of pigeonpea were more in the presence of marginal crops. This study clearly indicates the importance of availability of floral resources for honey bees under extensive insecticides application

Fungicides and herbicides impact many facets of honey bee colony health in a migratory beekeeping operation

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The detrimental effects of pesticides on honey bees is a major concern to commercial beekeepers. Sublethal effects of pesticides have been extensively evaluated in controlled lab studies and pesticide exposure in wax, pollen, and honey has been convincingly documented. This research is a facet of a larger 2-year study in a U.S. migratory beekeeping operation to apply epidemiological models to explain honey bee colony mortality. We evaluated pesticide residues in stored pollen and related these to metrics of colony health such as adult bee population, area of sealed brood, stored nectar and pollen, honey production, Varroa infestation, physiological stress markers, pathogen levels, and ultimately colony survival. The results show that pesticides may affect many aspects of colony health and survival, suggesting that multiple outcomes could be used to accurately evaluate the impact of pesticides under field-realistic conditions. Despite low acute toxicity, the impacts of fungicides and herbicides indicate future research should focus on these pesticide classes. These data link landscape use to pesticide exposure and ultimately colony health outcomes, which is critical for understanding complex and dynamic interactions that may cause high annual mortality in migratory beekeeping operations.

Can application time limit fungicide exposure to honey bees (Hymenoptera: Apidae) in almonds?

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Two fungicides, cyprodinil (cyp) and propiconazole (prop) are applied to almond (Prunus dulcis) blooms to arrest or stop growth of fungal pests. Application time of fungicides has the potential to disrupt pollination services by honey bees (*Apis mellifera*, L.) by alteration of behavior or physiology. This field study was undertaken to determine if evening or morning fungicide spray times affected honey bee foraging behavior differently. Prop and cyp were sprayed at maximal allowable label rates in separate corners of an almond orchard in bloom at evening (6pm) and morning (7am). Fungicide concentrations in pollen were analyzed from anthers and hives before and after spray times. Foraging behavior of honey bees was measured before and after spray times. The results show that fungicide concentration in anther pollen spikes after spray events as expected and pollen collected by bees showed accumulating levels of fungicides relative to distance and time. Forager presence in the spray areas decreased significantly over time in contrast to constant levels of returning foragers at the hives. Spray time affected exposure, and honey bees showed sensitivity to spray areas through decreased foraging in those areas. Concentrations of the fungicides in anthers were on the order of 104 ppb after sprays while concentrations in pollen collected at hive entranceways were on the order of 102 ppb in spray areas.

Field Analysis of Actual Exposure of Honey Bees to Neonicotinoids in Various Landscape Scenarios

T. Lawrence, A. Felsot, W. Sheppard
Washington State University - Entomology, Pullman, USA

Neonicotinoids have lethal and sublethal effects on bees, based on analysis of field-realistic levels. Honey bees forage over large areas with a broad diversity of floral sources, raising the question, what actual level of exposure to neonicotinoids does a colony experience from a given area? This is an essential question for beekeepers, citizens, and the political jurisdictions seeking to restrict the use of some or all of the 617 products containing neonicotinoids in the State of Washington. Neonicotinoids are the most widely used insecticide of the five major chemical classes and unwarranted restrictions on their use may present a significant disruption to agriculture. However, the loss of pollinators due to exposure to these products may also cause a significant disruption to agriculture and our food supply. To address this question, we studied the aggregate of floral sources in which bees forage. The levels of neonicotinoids in three different landscape scenarios in Washington State: urban, rural non-agricultural, and rural agricultural areas were sampled to determine the actual honey bee colony exposure to neonicotinoid insecticides from pollen foraging. We surveyed one hundred and forty-nine apiaries, ranging in size from one to hundreds of colonies, managed by commercial, sideline (semi-commercial), and hobby beekeepers. Our study evaluated residues in/on wax and beebread (stored pollen in the hive) for the neonicotinoid insecticides imidacloprid and its olefin metabolite and the active ingredients clothianidin, thiamethoxam, and dinotefuran. Beebread and comb wax collected from hives in agricultural landscapes were more likely to have detectable residues of thiamethoxam and clothianidin than that collected from hives in rural or urban areas (50% of samples vs. <10%). The maximum neonicotinoid residues detected in either wax or beebread was 3.9 ppb imidacloprid. A probabilistic risk assessment was conducted on the residues recovered from the beebread samples. The calculated 95th percentile risk quotient using 5 ppb as a dietary no observable effects concentration (NOAEC) for colony health parameters based on residues in beebread was less than 1.0. Thus, probabilistic risk characterization suggests a low potential for adverse effects on bee behavior or colony health from actual exposure levels of neonicotinoids in field conditions.

Sublethal effects of imidacloprid on targeting muscle and ribosomal protein related genes in the honey bee *Apis mellifera L.*

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A sublethal concentration of imidacloprid can cause chronic toxicity in bees and can impact the behavior of honey bees. The nectar- and water-collecting, and climbing abilities of bees are crucial to the survival of the bees and the execution of responsibilities in bee colonies. Besides behavioral impact, data on the molecular mechanisms underlying the toxicity of imidacloprid, especially by the way of RNA-seq at the transcriptomic level, are limited. We treated *Apis mellifera L.* with sublethal concentrations of imidacloprid (0.1, 1 and 10 ppb) and determined the effect on behaviors and the transcriptomic changes. The sublethal concentrations of imidacloprid had a limited impact on the survival and syrup consumption of bees, but caused a significant increase in water consumption. Moreover, the climbing ability was significantly impaired by 10 ppb imidacloprid at 8 d. In the RNA-seq analysis, gene ontology (GO) term enrichment indicated a significant down-regulation of muscle-related genes, which might contribute to the impairment in climbing ability of bees. The enriched GO terms were attributed to the up-regulated ribosomal protein genes. Considering the ribosomal and extra-ribosomal functions of the ribosomal proteins, we hypothesized that imidacloprid also causes cell dysfunction. Our findings further enhance the understanding of imidacloprid sublethal toxicity.

BEE HEALTH

10 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

IMPACT OF PESTS, PARASITES AND PATHOGENS ON BEES I ROOM 517D

[Lead-off] Honey bee social immunity towards varroa through Varroa Sensitive Hygiene (VSH)

F. Mondet
INRA UR 406, Avignon, FRANCE

Kin recognition and assessment of nestmate health-status are central to the collective defenses that social insects have evolved to counter pathogenic threats. One of the most important social immunity defenses in the honey bee, *Apis mellifera*, is hygienic behavior, which involves the detection and subsequent removal of abnormal or diseased brood. When bees specifically detect brood infested by the parasite Varroa destructor, the behavior is known as Varroa Sensitive Hygiene (VSH).
Evidence suggests that hygienic behavior and VSH in particular involve chemical communication, but which individuals from the colony participate in the behavior and how those bees choose which brood to target and remove is still partially unknown. Using chemical ecology techniques, we searched for signals that may be at the basis of VSH behavior initiation, as well as for signatures of VSH performers.
Our findings improve our understanding of the mechanisms of VSH behavior and highlight the importance of chemical communication as well as the role of the peripheral nervous system in the ability of honey bee colonies to fight infestations by varroa. Current efforts focus on the applied potential of these findings to develop methods to phenotype colonies for the VSH trait, with the perspective of promoting the selection of varroa resistant honey bee populations.

Honey bee parasite transmission via hive products

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The health of European honey bee subspecies, *A. mellifera*, has recently received considerable attention due to major losses of managed colonies at a global scale. Parasites can play a role for the mortality of colonies such as the microsporidian Nosema ceranae and the trypanosome Crithidia mellificae and the trade of hive products could potentially promote the spread of these parasites. However, few empirical data are available on hive products acting as matrices for parasite transmission. Here, we performed controlled laboratory hoarding cage assays to investigate whether the hive products honey, pollen and wax have a differential potential to promote the spread of N. ceranae and C. mellificae. For this purpose, hive products were spiked with three different concentrations of the parasites. The results show that N. ceranae can be successfully transmitted by all tested products resulting in high infection levels of the exposed bees. However, C. mellificae was only successfully transmitted by honey. This study shows that trade in bee products can significantly contribute to the spread of these pathogens.

A comparison of irradiation and storage time effects on transmission of waxborne viruses in experimentally reared honey bees

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Waxborne viruses have a demonstrated effect on the health of honey bees and can affect colony productivity if bees are hived on infected comb from dead colonies. Irradiation has been shown to reduce detectable levels of waxborne viruses and also influence virus levels in bees living on this comb. Additionally, storage time significantly reduces viruses on wax after 30 days, but the impact of these reduced virus loads on comb on infection rates in colonies has not been tested . This study explores the relationship between viral loads of Black Queen Cell Virus (BQCV), Deformed Wing Virus (DWV), Israeli Acute Paralysis Virus (IAPV) and Sacbrood Virus (SBV) in honey bees and virus levels on frames they were reared on, and effectiveness of mitigation treatments for reducing waxborne viruses. Frames for experiments were standardized by having bees draw out foundation for five days in either low virus (sourced from New Zealand colonies; n=30) or high virus (sourced from Varroa rearing colonies; n=30) colonies. Drawn frames were then assigned to three treatment groups: (1) frames stored for 35 days (n=20), (2) treated by ~40 kGy e-beam irradiation (n=20), (3) or a negative control (no storage or irradiation; n=20). Treatment frames were randomly and blinded assigned to rearing colonies, one frame per colony. Rearing colonies were 5-frame units, composed of low virus workers (n=60) sourced from New Zealand stock. Fifty-four of the rearing colonies were headed by New Zealand queens with one additional Kona queen added per treatment group. Initial virus levels were tested both from adult workers in each rearing colony and from wax of each treatment frame before introduction into rearing colonies. The first-laid cohort of bees on experimental frames were sampled as purple-eyed pupae, 10 day-old-adults, and 22 day-old-adults. Levels of BQCV, DWV-A, DWV-B, IAPV, and SBV were quantified using RT-qPCR in bees at each age and compared among wax source and mitigation treatment. The study will identify the viruses commonly transmitted in colonies via wax and the relative effectiveness of storage time and e-beam irradiation on each of these viruses.

Queen Health - Evaluation of Imported and Local Queens in Canada

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Honey bee queen health and performance is a crucial determinant of colony health and productivity. In Canada, a large number of queens are imported, mainly from northern California and Hawaii, although queens may also be imported from Australia, New Zealand, and Chile. In 2017, 225,000 queens valued at 7 million dollars were imported. According to beekeepers, the performance of local and imported queens is variable and queen replacements are needed more often than in the past. We therefore investigated the health status of freshly mated queens, both locally produced and imported, by analysis of pathogens and the quantity and viability of the sperm the queen carried. Sperm quantity and viability are crucial for the queen to efficiently lay fertilized eggs and head a commercial beekeeper operation. Comparative analysis of local and imported queens showed that imported queens had lower and more variable sperm viability as assessed in the laboratory by differential fluorescence staining.
Potential causes of low sperm quality will be discussed including challenges during mating or transport. Further, analysis of established queens obtained from weak colonies showed that their sperm viability was 60%, which may explain poor colony performance. Higher viral abundance and viral titers were also found in these queens and may be affecting their performance.
Pathogen analysis of freshly mated queen and their shipment attendants included viruses, Nosema spp, and Trypanosomes. Freshly mated local and imported queens carry low levels of Deformed Wing Virus, although queen attendants tested positive for Nosema spp. as well as the new Trypanosomatid parasites. A general recommendation resulting from this assessment is to re-cage the queens or remove attendants before queen introduction.

Do honey bees die over winter from the flu?

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In this presentation we report about a metagenomic study of honey bees performed on pooled samples of adult Belgian bees enriched for viral particles according to the NetoVIR protocol. The study revealed several novel bee viruses, one of which was member of the family of Orthomyxoviridae. These are RNA viruses that include seven genera, 3 of which are of importance for human as they contain the different Influenza viruses (Influenza virus A to C), better known for the flu pandemics they can cause (Influenza virus A). The virus that we found was the closest related to the members of the Thogotovirus genus, which represents viral species that infect insects (mosquitos) and ticks. To our knowledge an Orthomyxo-like virus has so far never been discovered in bees. We performed RT-qPCR to measure the prevalence of this new virus and found relative low presence in the Belgian honey bee population, though there was a significant ($p = 0.0246$) association with winter losses. The absolute abundances of these viral particles in bees from infected colonies seems to be high. We examined different developmental stages of the honey bee in an infected colony and could demonstrated the presence of the Orthomyxo-like virus from egg to adult bee. All tagmata of the adult bee are affected suggesting a systemic infection. We therefore developed a procedure for transmission electron microscopy (TEM) to capture the hemocytes after citrate treatment (to avoid clotting) based on low melting temperature agarose. The TEM images revealed the typical endocytosis of viral particles for cell entry, amplification of the virus in the Golgi-apparatus and budding off at the cell membrane of new virus particles for release. Infected hemocytes show severe cytopathological signs, which is fully in line with the major impact on bee survival that we observed.

Geographic mosaic and host specificity of Varroa destructor mites infesting Western and Eastern honeybees

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The ectoparasitic mite, Varroa destructor, shifted hosts from Eastern honeybees, *Apis cerana*, to Western honeybees, *Apis mellifera*. Even though it is known that genotypic diversity and reproductive performance of V. destructor mites infesting the different honeybee species can differ significantly, large scale studies comparing sympatric mites in both host species remain scarce. Here, we investigated the mites of the V. destructor Korea haplotype, which gave rise to the globally invasive mite lineage (K1 haplotype), in a large-scale survey from North to South China (*A. cerana*: N=102 colonies, N=599 mites; *A. mellifera*: N=25 colonies, N=373 mites). Our results confirm a higher genetic diversity of the K1 haplotype in *A. cerana*, as much as four mite variations in one original host colony, compared to *A. mellifera* and new mitochondrial haplogroups and variants were found. Surprisingly, the mites from *A. cerana* in East Central China are genetically closest to mites in *A. mellifera*. Nuclear DNA suggests that local mite populations can vary in their reproductive abilities on *A. cerana* and *A. mellifera*, thereby supporting the geographic mosaic of co-evolution. Furthermore, V. destructor K1 mites from *A. cerana* populations were with few exceptions genetically distinct from the mites in *A. mellifera* colonies, suggesting that the variants, which can successfully shift hosts are rare. Most likely, an investment towards mite reproduction in host worker brood is strongly selected against in *A. cerana* and favored in *A. mellifera*. The differences between mites from the same K1 haplotype highlight the need to further investigate these V. destructor mites in their original and new hosts to better understand this major threat to *A. mellifera* worldwide.

Flowers as dirty doorknobs: Virus transmission through shared flowers depends on floral diversity

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Evidence is mounting that RNA viruses, likely originating from honeybees, have spilled over into wild bee communities. While transmission of disease between bee species likely occurs through shared flowers, only two published studies have directly examined this floral transmission route and no study has yet examined their role in RNA virus dissemination. Our previous work has shown that flowers can harbor viruses with prevalence varying across floral species. This indicates that floral diversity, density and their patch distributions may influence transmission dynamics. In this study, we experimentally demonstrate how flowers may be facilitating the spillover of Deformed Wing Virus (DWV) from honey bees into bumble bee communities. In addition, using an agent-based mathematical model, we examine how flower communities influence RNA virus spread in bumble bees. Clean bumble bees were allowed to forage on hand inoculated and naturally infected red clover. We measured the viral load that can be picked up by a bee from a flower as a function of foraging time determined the dose curve using RT-qPCR. We found that DWV can be picked up by bumble bees from field realistic hand-inoculated flowers and flowers visited by infected honeybees. Foraging time was positively correlated with viral load and 1 million genome copies of DWV caused an infection with ~30% prevalence. Using these parameters, we constructed a cellular automata model on a network topology with flower patches as nodes and distances between patches as edges. Optimal foraging theory was used to model individuals' decisions to move across the network. Infected bees were able to infect flowers depending on the flowering species' harboring potential γ. Uninfected bees picked up viruses from infected flowers with a probability, β. Birth and death rates for all agents were taken from the literature. Preliminary model analysis shows that the simulation with the primary floral transmission route closely match DWV prevalence in our field data. In addition, in cases where flowers with low viral harboring potential are numerous, we found a dilution effect on transmission. The demonstration of this transmission route and the model findings may be used to more accurately inform management recommendations.

SYMPOSIUM — 13.00-15.00
IMPACT OF PESTS, PARASITES AND PATHOGENS ON BEES II — ROOM 517D

RNA virus spillover from managed honeybees (*Apis mellifera*) to wild Bumblebees (Bombus spp.)

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RNA viruses, once considered specific to honey bees, are suspected of spilling over from managed honeybees into wild *Bumblebee* populations through shared floral resources. In a field study, we examined if RNA viruses spillover from managed honeybees, the extent to which viruses are replicating within *Bumblebee*s, and the role of flowers in transmission. Prevalence and active infections of deformed wing virus (DWV) were higher in *Bumblebee*s collected near apiaries and when neighboring honeybees had high infection levels. We found no DWV in *Bumblebee*s where honeybees were absent. The prevalence of black queen cell virus (BQCV) was also higher in *Bumblebee*s collected near apiaries. Furthermore, we detected viruses on 18% of flowers, all of which were collected within apiaries. In controlled experiments using captive colonies in flight cages, we demonstrated that honey bees leave viruses on flowers but not equally across plant species. Our results corroborate the hypothesis that viruses are spilling over from managed honeybees to wild *Bumblebee*s and that flowers may be an important route for transmission. Our results also suggest that transmission via flowers may be mediated by differences in virus ecology, floral morphology, and/or pollinator behavior.

In Search of a Good Meal: Varroa destructor regularly switches between adult bee hosts to feed

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Varroa destructor, a parasite of the honey bee, *Apis mellifera*, has devastated honey bee populations globally, damaging its host in all life stages and vectoring viruses. The majority of varroa in a colony are found reproducing inside brood cells. Outside of the brood cells, varroa are considered by beekeepers to be in their phoretic phase. Phoretic strictly refers to a parasite using a host for transport and is not appropriate if the parasite is feeding. We found that phoresy does not accurately describe the behavior of varroa on an adult bee host. Using a combination of marking and fluorescent techniques, we tracked the movement of individual varroa and recorded their feeding behaviors on individual bees within a population of bees. Varroa regularly switched adult bee hosts and entered a feeding position between the abdominal tergites. Individual hosts and varroa were recollected and examined under fluorescent microscopy for proof of feeding. We

were able to track individual varroa, and to verify feeding on a bee host using our novel methods. High and low frequency rates of host switching and feeding were seen across trials. By manipulating the age of the adult bee host, the rate of host switching was modulated. Over the length of the 15 day trials, the frequency of host switching and feeding decreased. Key feeding behaviors and the role they play in vector capacity models will be discussed to illustrate how they impact the transmission of honey bee viruses.

Susceptibility to viral infection in honey bees driven by genotypic differences in tolerance and resistance

M. Simone-Finstrom
USDA-ARS, Baton Rouge, USA

The ectoparasitic mite Varroa destructor is the biggest threat affecting honey bee health in large part because of the viruses that mites vector while feeding during reproduction and development on honey bee pupae. Deformed wing virus (DWV), in particular, has been associated with colony losses. Because of the significance of Varroa-DWV dynamics, there has been much interest in the relationship between colony mite infestation and viral prevalence. In a few cases, it has been noted that colonies that have natural resistance mechanisms against Varroa have lower incidences of DWV infection. However in other populations mite-resistance seems to be correlated with tolerance to DWV, meaning that mite-resistant colonies survive with high levels of DWV and exhibit fewer symptoms. To clarify whether resistance (maintain low viral titers despite infection) or tolerance (high survival, no symptoms with high virus) to DWV appears to be driving these differential effects, a series of experiments have been conducted. 1) Pupae from single-drone inseminated queens were injected with low, moderate or high levels of a DWV inoculum and followed through emergence to examine survival and expression of symptoms. A subset of these pupae were analyzed from putatively resistant and susceptible colonies to determine differences in viral titer and antiviral responses as a way to detect whether the colonies exhibit resistance or tolerance to DWV. Additionally, pupae from the same colonies that displayed differential symptom development after injection with the same dose were examined to understand possible individual differences in tolerance or resistance to viral infection. 2) Newly emerged bees collected from 3 mite-resistant stocks (Russian, Pol-line and Saskatraz) and 2 mite-susceptible stocks (Italian and Carniolan) were injected with a DWV inoculum. Bees were subsampled over time to determine how bee genotype influences rate of dissemination of the virus throughout an individual by analyzing viral load in different. Results suggest that tolerance may be the major factor explaining genotypic differences in susceptibility and development of symptoms in pupae. Further, sufficient variation exists within a stock and across stocks with some stocks exhibiting greater variation than others indicating that tests for viral susceptibility could be valuable for breeding programs.

Understanding the Tropilaelaps Mite, An Emerging Threat to Western Apiculture: Analysis of Biology, Behavior, and Life Cycle

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Tropilaelaps mercedesae is a rapidly emerging threat to western apiculture. The recent, dramatic expansion of Tropilaelaps' host range and concomitant host shift to *Apis mellifera* has lead the World Organization for Animal Health (OIE) to classify Tropilaelaps as an agent capable of causing "considerable global economic injury". However, research progress has lagged far behind the perceived need. Fundamental questions as to how these parasites interact with their host below the capping remain open questions. To better understand the relationship of this parasite and its host, we placed several transparent, plastic cells into *A. mellifera* brood frames. Tropilaelaps were allowed to invade cells naturally, cells were capped by nurse bees, and the behaviors of the mites were recorded using a high-resolution video camera. Behaviors were coded and analyzed through the duration of the capped period. Frequency and duration of feeding events was noted in addition to preferred and avoided regions of the developing host. Mites appear to feed from a variety of locations throughout the development of the host unlike Varroa. Further, novel behaviors were observed and described. These findings provide us with a greater understanding of the stage of the mite's lifecycle where it most directly impacts its host, providing us with information can that be used to target vulnerabilities in the mite's lifecycle that can be targeted for the development of much needed remediation strategies.

Pathogenic Associations with Winter Colony Loss in Canada

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Canadian beekeepers report that high pathogen/parasite infestation levels, poor queen quality and severe weather conditions are the leading causes of elevated wintering losses. In order to replenish annual losses or maintain their operations, beekeepers in Canada face a unique and difficult situation for purchasing new queens or package bees. Scarce local supply drives local producers to import approximately 300,000 queens and packages each year, predominantly from foreign sources. This large-scale importation of stock may contribute to the introduction of undesirable pathogens or genetics, and supply bees that have not been selected to survive and prosper in northern temperate climates, thereby influencing wintering success.

Honey bees act as a host for a multitude of pathogens and parasites. Nevertheless, the interactive effects that many of these pathogens, endoparasites and ectoparasites have on colony wintering success remains poorly understood. In order to better understand these interrelationships, we studied colony health and wintering success as a part of an ongoing national-scale study. In 2016 and 2017, we sampled 1025 and 520 colonies, respectively, across five Canadian provinces. During each experimental year (May through April), we collected pre-winter phenotypic data (fall colony weight and cluster size), and samples for pathogen analysis (Nosema spp., *Lotmaria passim*, DWV-A, DWV-B, BQCV, SBV, and phoretic loads of Varroa destructor) from colonies in all locations to investigate the main drivers of colony winter mortality. We were also able to study colonies wintered outdoors, as well as those wintered inside specialized wintering facilities. Although winter mortality was statistically similar between 2016 and 2017 (15% and 13%), preliminary results indicate that there is a difference between years with respect to the impact that individual pathogens and colony phenotypes have on colony winter survival. Nevertheless, in both experimental years, levels of DWV-A, fall phoretic mite loads, as well as fall colony weights and cluster sizes exhibited significant influences on colony winter survival outcomes. Additionally, we will illustrate the distribution and abundance of colony pathogens across honey bee populations in Canada.

Interesting interactions? Effects of parasitic mites and neonicotinoid insecticides on honey bee food glands

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The ectoparasitic mite Varroa destructor and neonicotinoid insecticides are known to independently cause lethal and sub-lethal effects in honey bees. Recent studies demonstrated negative effects of both stressors on nursing worker hypopharyngeal glands (HPGs). However, little is known about potential combined effects on HPGs. Because HPGs are crucial to producing brood food that nourishes all honey bee types (queen, worker, drone), we examined the effects of simultaneous neonicotinoid and V. destructor exposure on HPGs by performing a fully crossed experimental design. Known age cohorts of worker honey bees were obtained from 24 colonies which were previously fed with pollen patties for 49 days. Half the colonies received patties that contained field-realistic concentrations of a model neonicotinoid (3.25 ppb thiamethoxam), whereas the other half received patties without the neonicotinoid. Workers from each colony were artificially emerged, assessed for V. destructor parasitism, and allocated to one of four treatments: 1. No stressors (controls), 2. V. destructor only, 3. Neonicotinoid only, and 4. Both neonicotinoid and V. destructor. We maintained workers in laboratory cages equipped with sugar syrup and pollen patties for 10 days, the typical age of nursing, and also monitored for mortality. Surviving workers were then decapitated for HPG examination. We hypothesized that individuals exposed to both stressors simultaneously would have smaller HPGs compared to other treatment groups. Contrary to our expectations, no significant difference in HPGs was observed in individuals exposed to both stressors compared to those exposed to V. destructor only ($p=0.699$); however, both treatment groups significantly differed from controls (p's<0.05). Since no effect was observed on HPGs when individuals were exposed to only neonicotinoids ($p=0.113$), our results suggest that V. destructor is the prime stressor and does not interact with neonicotinoids to negatively affect HPGs. They further emphasize the importance of proper V. destructor management in order to improve honey bee health.

The impact of viruses on honey bees at the individual and cellular levels

M.F. Flenniken [1], K. Daughenbaugh [1,3], A. Mcmenamin [1,2,3], L. Brutscher [3], F. Parekh [2,3], M. Flennken [1,2,3]
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Honey bee colony losses are influenced by multiple abiotic and biotic factors, including viruses. To better understand the effects of RNA viruses on honey bee health at the individual level, we infected bees with a model virus (Sindbis-GFP) in the presence or absence of dsRNA. In honey bees, dsRNA is the substrate for sequence-specific RNA interference-mediated antiviral defense and is a trigger of sequence-independent (non-specific) antiviral responses. Transcriptome sequencing identified more than 200 differentially expressed genes, including genes in the RNAi, Toll, Imd, JAK-STAT, and heat shock response pathways, and many uncharacterized genes. To confirm the virus limiting role of two genes (i.e., the RNAi endonuclease dicer and an uncharacterized gene MF116383) in honey bees, we utilized RNAi to reduce their expression in vivo and determined that virus abundance increased, supporting their involvement in antiviral defense. To evaluate the role of the heat shock stress response in antiviral defense, bees were heat stressed post-virus infection and virus abundance and gene expression were assessed. Heat stressed honey bees had reduced levels of virus infection compared to controls and the expression of one small heat shock protein (Hsp) (protein-lethal(2)essential for life-like) was increased in the context of heat stress and virus infection.
To determine if these genes are universally associated with antiviral defense, honey bees were infected with another model virus (flock house virus) or deformed wing virus and gene expression over the course of infection was assessed. Dicer expression was greater in bees infected with either FHV or Sindbis-GFP compared to mock infected bees, but differential expression was not observed in deformed wing virus infected bees.
To further investigate honey bee antiviral defense mechanisms and elucidate the function of key genes (dicer, ago-2, MF116383, and Hsp encoding genes) at the cellular level, primary honey bee larval hemocytes were transfected with dsRNA or infected with Lake Sinai virus 2 (LSV2). These studies indicate that MF116383 and Hsps mediate dsRNA detection and that MF116383 is involved in limiting

LSV2 infection. Together, these results further our understanding of honey bee antiviral defense, particularly dsRNA-mediated antiviral responses, at both the individual bee and cellular levels.

Impacts of Nosema ceranae and Lotmaria passim on honey bee physiology and behaviour

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Nosema ceranae and *Lotmaria passim* are two commonly encountered, emerging eukaryotic parasites of the honey bee (*Apis mellifera L.*). Nosema ceranae is a midgut-infecting microsporidian that negatively impacts honey bee health at both the individual and colony level. *Lotmaria passim* is a globally prevalent, recently characterized trypanosomatid. Given its emergent nature, little is known regarding how L. passim affects honey bee physiology and behaviour, and despite the frequency with which we detect mixed infections of N. ceranae and L. passim in Canadian and imported stock, it is unknown how these two parasites interact to affect honey bees. We investigated the impact of both parasites (single and mixed infections) on honey bee mortality, humoral defense responses, and behaviour. Results of a mortality experiment performed in 2018 suggest L. passim is less virulent than N. ceranae alone, with individuals inoculated with only L. passim surviving 10.4 days longer than those inoculated with only N. ceranae. Surprisingly, infections of L. passim and N. ceranae in combination appear less virulent than N. ceranae alone. Workers inoculated with both parasites survived 0.75 days longer than those inoculated with just N. ceranae. We will also discuss the impact of single and mixed infections on the temporal expression of five antimicrobial peptides, and on honey bee behaviour. Our goal is to provide the beekeeping community with much-needed knowledge regarding the impacts of two emerging parasites on honey bee health.

SYMPOSIUM 15.30-17.30
BREEDING FOR DISEASE / MITE RESISTANCE I ROOM 517D

[Lead-off] Breeding Honey Bees for Disease and Mite Resistance

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Since 2006, most research has focused on understanding the impact of individual stressors and their interactions leading to honey bee colony mortality. Our approach instead focuses on colony health and survivorship through honey bees' natural and collective behavioral defenses called social immunity. Our goal is for the beekeeping industry to propagate bee stocks that demonstrate multiple traits of resistance against pathogens and parasites, and thus reduce the need for human intervention. One form of honey bee social immunity is hygienic behavior; another is the collection and formation of resins into a propolis envelope within the hive that acts as an important antimicrobial layer. Colonies that display hygienic behavior actively reduce pathogen and Varroa loads. The presence of a propolis envelope within colonies benefits individual immunity and reduces some pathogen loads in the colony. Based on these findings from our previous research, we have started a comprehensive breeding program that combines mechanisms of social immunity. From colonies that winter well in our northern climate, are gentle and good honey producers, we are selecting for hygienic behavior using new chemical assays, high propolis collection, and low Varroa mite growth over the season. We will use a combination of instrumental insemination and natural mating in this breeding program, which we anticipate will take 3-5 years to achieve demonstrable results.

Successful breeding for honeybee virus resistance in Belgium based on the 'lack of vertical transmission' trait

D. De Graaf [1], D. Laget [1], M. Brunain [1], L. De Smet [1], E. Danneels [1], B. Rotthier [1], P. Demaeght [1], R.F. Veerkamp [2], E.W. Brascamp [2]
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In 2012 we introduced the selection criterion 'lack of vertical transmission' in our honeybee breeding program. The trait was estimated by determining the virus load in a sample of 10 eggs. We hypothesized that queens capable of clearing a virus infection in the ovaria, resulting in the deposition of virus-free eggs, will inherit this trait to their progeny leading also to a better control of viral infection at colony level. Egg samples were analyzed for the presence of ABPV, BQCV, DWV and SBV by RT-PCR. After 6 years of breeding efforts, we found a remarkable decrease in the total number of virus-positive samples, which dropped in 2018 for the first time under 10% (7.02%) for DWV, whereas in the early years this was up to 44.12%. For the subgroup of queens that were the descents from virus-negative mother queens, the DWV infection was even lower (6.06%). With a data set of 694 records in hands, representing 2776 virus analyzes, we recently performed a statistical analysis in order to determine the heritability of this trait. One of the models used gave a statistical substantiated heritability h^2 of 0.19 ± 0.18. The large estimation error can be expected because of the limited size of the dataset. The heritability value h^2 confirms, however, that variation of this new trait can partially be explained by genetic effects. We have identified several pedigrees with 3 consecutive generations of queens with no vertical transmission of viruses and the trait seems to be inherited both from mother and father. Next bee season we will continue our efforts and further investigate what it means in terms of virus load of different tissues of the queen and the spread of the disease in the colony. The discovery that 'lack of vertical transmission' is inherited, opens new opportunities

for breeding programs all over the world as it will strengthen the resilience of the honeybee colonies and allows to expand our focus in addition to the Varroa-mites, also to the viruses they transmit by vectoring.

Evaluation of characters for testing on varroa resistance

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Varroa destructor is probably the most serious cause of European honey bee (*Apis mellifera*) losses on a global level. However, the presence of untreated survivor populations in many different regions support the idea of selecting resistant stock. Recently, the European Commission requested a study on the present status of breeding activities regarding varroa resistence and its commercial relevance for the European apicultural sector. A preliminary evaluation of interviews with about 50 breeding experts around the world resulted in an interesting of favored selection characters, including differences in mite infestation, brood hygiene behavior (HYG), brood uncapping/recapping (REC) and suppressed mite reproduction (SMR).
Those characters have been investigated in A.m.carnica breeding populations in Austria, Croatia and Germany during 2016 and 2017 under standardized performance test conditions. Repeated bee samples were used to estimate the infestation level of colonies. Repeated pin tests involving 50 brood cells each with an evaluation of opened cells after 8 hours were applied to estimate HYG behavior. On average 260 brood cells resp. 23.6 single infested cells were investigated per hive to evaluate REC and SMR behavior according to a standardized protocol.
The 294 tested colonies showed on average (minimum to maximum) an infestation level of 3.4 (0.0 to 26.8) mites per 10 g of bees, a HYG level (rate of opened cells) of 85.7 (14.0 to 100) %, REC of infested cells of 40.5 (0 to 100) % and SMR of 27.5 (0.0 to 90.0) %. A partial correlation analysis with country and season as controlling variables revealed significant correlations between SMR and REC ($r=0.248$, $p=0.000$), SMR and HYG ($r=0.133$, $p=0.040$), HYG and bee infestation ($r=-0.239$, $p=0.000$) and finally HYG and REC trait ($r=0.194$, $p=0,003$).
Those weak but highly significant correlations across three independent Carnica populations during two seasons underline the general relevance of those parameters. HYG, SMR and REC seem to be related characters that negatively affect mite population development of the colonies. They can thus be recommended as parameters for the selection on Varroa resistance although their contribution to the total variability of mite infestation seems to be limited.

Naturally selected Varroa-resistance behaviors in unmanaged survivor bees living in the Arnot Forest, NY, USA

D. Peck, T. Seeley
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Mites of the genus Varroa are specialist parasites of honey bees (genus Apis.) A recent (c. 100 years) host shift event enabled mites specialized to exploit eastern honey bees (*Apis cerana*) to infest colonies of western honey bees (*Apis mellifera*) and speciate into Varroa destructor, causing devastating economic and ecological harm to both managed and wild bee populations globally. We investigated how a population of European-derived western honey bees surviving, without human management or mite treatment, in the Arnot Forest near Ithaca, New York is able to persist despite infestation with these parasites. We describe the presence of multiple mite-resistance traits, including high levels of brood hygienic behavior and mite-grooming behavior, which may synergize to significantly limit the growth of Varroa destructor populations in these colonies. Comparing our observed levels of mite-resistance phenotypes to historic measurements of similar unselected populations of honey bees, we infer baseline levels of these mite-resistance traits. Our data are consistent with the hypothesis that the Arnot Forest survivor bees have rapidly evolved to increase expression of multiple, apparently independent, mite-resistance behaviors conferring partial resistance to these parasites.

Apis mellifera resistance to Varroa destructor across continents

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Varroa destructor is the major biotic threat to the Western honey bees *Apis mellifera*. Most colonies infested with this ectoparasitic mite cannot survive if they are not treated with acaricides. Fortunately, some *A. mellifera* populations present natural defense mechanisms to overcome the infestation by mites and/or by varroa-transmitted viruses and can survive extended periods without the need for treatments. This is the case of Africanized honey bees inhabiting the northeast part of Uruguay, and a population of mite-surviving colonies in Avignon, France.
This study aimed to characterize possible traits associated with the natural survival of these populations.
Experimental apiaries were installed at the beginning of spring in Avignon (France) and Treinta y Tres (Uruguay). Each apiary was composed of colonies that survived for over three years without acaricide treatment, and control colonies from a population that normally requires

treatments to survive varroa infestation. In both regions, the same protocol was used to assess different honey bee behaviors (VSH and grooming), brood resistance traits (SMR), the effect of the mite and the prevalence and titers of the major *A. mellifera* viruses.
Our results show that different behaviors are contributing to colony survival in the two surviving honeybee populations. Besides that, we detected genetic differences in the mites infesting the control and surviving colonies in France that matched phenotypic differences in the colony response to infestation. Moreover, the viruses infecting the surviving and control colonies differed significantly in the two countries. These exciting findings will help improving current breeding strategies against the honeybee's enemy number one.

Brood signals as the basis for an improved selection tool for hygienic honey bees

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Varroa mites remain one of the most important biological threats to honey bee health. Selective breeding for naturally occurring social immunity traits has emerged as one sustainable approach to suppressing the mites' impact on honey bees. Based on our finding that brood genotype influences the efficiency of hygienic behavior, we have identified several cuticular hydrocarbons of honey bee brood that are associated with mite parasitization and related disease. These compounds may thus represent signals capable of inducing hygienic behavior in nurses. In follow-up experiments, the application of synthetic versions of several of these compounds triggered uncapping and removal behavior, confirming their role in hygienic behavior. After some optimization, we have formulated a mixture of these compounds that effectively elicits hygienic behavior, and we can now report the relationship between colony response to a spray assay of the mixture and colony health indicators, such as mite levels. Thus, we discuss the applicability of this assay as a user-friendly and efficient selection tool that can be incorporated into a wide range of honey bee breeding programs.

Environmental factors influencing Varroa Sensitive Hygiene trait in the honey bee

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Varroa resistance can be defined as the ability of honey bee colonies to survive the parasite for several years in the absence of any treatment against the mite. Long-term survival of untreated *Apis mellifera* populations has been reported in the US and Europe. The ability of honey bee colonies to survive varroa mite infestations has been associated with the development of Varroa Sensitive Hygiene behavior (VSH). Resistant colonies are able to detect the presence of varroa through the cap of developing brood cells and to remove parasitized brood and the mites. To improve breeding programs for varroa resistance, detailed knowledge of the mechanisms that enable bees to survive mite infestation and environmental conditions that can influence the expression and evaluation of VSH behavior are needed. This study aims at evaluating the influence of the colony population dynamics, the varroa density in colonies, the task allocation strategy and the food supply on the ability of colonies to express the VSH trait. Such efforts are particularly important to standardize testing in different locations, a feature that is essential to ensure the success of breeding efforts

BEE HEALTH

11 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

BREEDING FOR DISEASE / MITE RESISTANCE II ROOM 517A

New accessible methods to facilitate honey bee collection & preservation for selective breeding programs based on proteomic markers

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Effective sample collection in the field is key for the success of honey bee (*Apis mellifera*) breeding programs based on proteomic markers. Samples are ideally snap frozen in the field using dry ice, shipped in large parcels containing an adequate amount of dry ice, and maintained frozen in the laboratory until further processing. However, this collection method presents multiple obstacles, including difficult access to dry ice for beekeepers and sample integrity loss during delayed shipping. To overcome these issues, and simplify sample collection while preserving sample integrity, two alternative preservation methods were investigated: (1) 70% ethanol, which is easily found in laboratories; (2) 70% isopropanol, which can be commonly bought in drug stores. Using an Agilent 6460 triple Quad LC/MS, we tested the presence and potential degradation though time of 22 published protein markers in honey bee antenna samples preserved under different conditions (70% ethanol and 70% isopropanol at room temperature in the dark), and analysed at different time points (7, 14, and 21 days after collection). Control samples were snap frozen in dry ice and kept at -72°C, and analysed at the same time points. After optimizing the protein extraction protocol, we successfully increased the number of detected markers in alcohol preserved samples (13 markers for 70% ethanol, and 9 markers for 70% isopropanol) up to 87% of the markers detected in the snap frozen samples (15 markers) for 21 days. These results indicate that beekeepers could use 70% ethanol and 70% isopropanol to collect bees, and send them to laboratories within three weeks. Further optimization of the extraction protocol will help improve marker detection. At length, we hope these changes will facilitate beekeepers' participation in breeding programs based on proteomic markers, and promote the development of such selective breading programs.

Genetic selection of the honeybee (*Apis mellifera L.*) in a northern climate

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Animal breeding, in combination with developments in agricultural technology, has made remarkable progress in increasing production of many domestic species. However, these important tools are poorly exploited in the honeybee (*Apis mellifera L.*) industry because of the complex genetic and reproductive features of the bee. In recent years, new mathematical approaches have allowed the application of statistical models in honey bee breeding programs and the use of breeding values to improve genetic selection. The aim of our project is to adapt statistical models currently used for breeding in the Dairy and Pig Industry in Quebec Canada for the genetic evaluation and performance monitoring of the honey bee. This project will: 1) identify honey bee performance traits with high heritability within the colonies of the Centre de recherche en sciences animales de Deschambault honey bee breeding program; 2) develop a breeding plan to improve the genetic potential and produce superior breeding stock and 3) measure the impact male selection on the breeding values of honey bee colonies. This novel approach will improve honey bee performance of traits relating to yield stability and sustainability of our northern climate beekeeping industry.

Disentangling the honey bee pathogen-web and its dynamics on colony productivity, health phenotypes and social immunity behaviours

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Pathogens and parasites have evolved to overwhelm and suppress their host's immune system, but their effects on honey bee colony productivity traits and pre- post-winter phenotypes are still largely unknown. As a defense mechanism, social insects have evolved remarkable behaviours at the social level (social immunity) to counter several pathogen/parasite challenges, which can reduce colony-level disease, and improve colony health. In 2016 and 2017, 1025 and 520 colonies, respectively, were sampled across Canada (British Columbia, Alberta, Manitoba, Ontario and Quebec). During each experimental year (May until April of the following year), we collected phenotypic data and samples for pathogen analysis from colonies in a standardized manner. We measured colony size and productivity (colony weight and cluster size, honey production, and sealed brood population), social immunity traits (hygienic behaviour, instantaneous

mite population growth and grooming behaviour), and quantified loads of gut parasites (Nosema spp., and *Lotmaria passim*), viruses (DWV-A, DWV-B, BQCV and SBV) and Varroa destructor. Our comprehensive goal is to examine: 1) correlations between pathogens and colony phenotypes; 2) dynamics of pathogens and parasites on colony phenotypes and productivity traits; and 3) effects of social immunity behaviours on colony pathogen load. To date, we have performed multiple correlation and regression analyses to assess the relationships between pathogen and colony-level phenotypes/traits/behaviours. Our preliminary results show that high fall phoretic mite loads and high levels of DWV-B were associated with low colony weights and small cluster sizes in the fall. We also found that increases in colony hygienic behaviour performance were associated with decreases in levels of DWV-A in spring-sampled bees. Furthermore, we discovered that cluster size and weights of colonies after winter were inversely associated with SBV and BQCV virus loads during spring, as well as with post-treatment mite levels the previous fall. Our goal is to further disentangle the inter-correlation of several important pathogens and phenotypes, so as to better understand drivers of colony health and productivity. This in turn will enable local queen producers to make informed management decisions when selecting and breeding colonies to be healthy, productive and well-adapted to the Canadian climate.

The Russian Honey Bee Program: An update since the stock's release in 2008

L. De Guzman
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The Russian honey bee (RHB) program released 17 lines that are resistant to Varroa destructor and Acarapis woodi and are good honey producers. Since 2008, the Russian Honeybee Breeders Association (RHBA) has been responsible for maintaining and selecting to improve the stock. We continue to use the stock for various behavioral studies including removal response towards mite-infested brood, grooming behavior, response to virus infection, and flight activities. RHB colonies were more hygienic towards mite-infested brood than Italian honey bees (IHB). Further, the RHB colonies that displayed the highest levels of hygiene also groomed Varroa longer. Regarding viral load, we found that both RHB pupae and their infesting mites had lower levels of Deformed Wing Virus and Chronic Bee Paralysis Virus than infested IHB and their corresponding Varroa, which may have helped RHB perform more flights which had longer flight durations. In 2016, the performance of Russian queens produced by the association (RHBAQ) was compared to those of commercial IHB, IHB mated with Russian drones (IHB-RUS) at our laboratory's mating yard, and two commercially advertised Russian queens that were not produced by the association (non-RHBA1 and non-RHBA2). This experiment was repeated in 2017. For both years of evaluation, we found significant differences among the five genotypes for bee population, mite population and colony survival. Overall, the non-RHBA1 colonies were the smallest, but not different from non-RHBA2. While IHB colonies displayed high susceptibility to Varroa, pure RHBAQ maintained their mite resistance. The IHB-RUS hybrids were intermediate in resistance to that of pure RHBAQ and IHB colonies. Mite population growth in the two commercially advertised non-RHBA was inconsistent suggesting that not all non-RHBA queens have equal mite resistance, which may also vary from year to year. Low mite numbers may have contributed to the increased survival of RHBAQ colonies. Overall, our results suggest that Russian queens produced by RHBA have maintained their mite resistance, which probably resulted from a systematic breeding plan and drone-sharing among members.

BeeStrong: towards a genomic tool for the selection of varroa resistant honey bees

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The mite Varroa destructor is known for causing devastating colony losses in the western honey bee, *Apis mellifera*. While chemical treatments are currently the norm in many countries, the most sustainable solution against this pest is to select and breed varroa-resistant honey bees. However, this solution is impeded by the difficulty to evaluate the ability of colonies to resist to the mite and the labour intensity of the currently existing methods.
The BeeStrong project aims at developing a genetic diagnosis tool for varroa resistance that will simplify selection for beekeepers and research facilities. To achieve this goal, phenotypic data have been collected from over 1500 colonies between 2016 and 2018 in France, Switzerland, USA, New Zealand, Luxembourg, Sweden and the Netherlands. The data consists in estimations of colony performance (ColEval method), phoretic mite infestation and evaluation of the suppressed mite reproduction (SMR) trait. Our dataset gives us new insights into the SMR trait and varroa resistance in general.

Local adaptation of naturally surviving bees to their sympatric mites?

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After host switches of parasites, coevolution is obviously lacking, which may lead to major impact on the novel susceptible hosts. This was the case, when ectoparasitic mites, Varroa destructor, switched hosts from Eastern to Western honey bees, *Apis mellifera*. Since beekeepers usually treat colonies, the *A. mellifera* honeybees cannot adapt. Nevertheless, untreated *A. mellifera* are known to survive for

>10 years by means of natural selection, mainly due to adaptive shifts in bee behaviour, e.g. recapping of brood cells. However, the bees are only one side of the Darwinian coin and possible local adaptations of the mites and bees have largely been ignored so far. Here, we examined possible local adaptations of Dutch surviving colonies to their sympatric mites. Artificial cross infestations experiments were conducted using mites and bees of local susceptible and surviving colonies. Recapping behaviours of the bees as well as reproductive parameters of the mites were investigated. The data will be presented and discussed.

Confirmation and Application of SNPs Related to Chalkbrood Resistance in Larvae of *Apis mellifera*

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Chalkbrood is a contagious fungal disease caused by Ascosphaera apis in honey bees (*Apis mellifera L.*), which causes colony population decrease, productivity declines and income decrease.. The resistant breeding is an effective and ecological solution to control chalkbrood currently. This study aimed to screen reliable single nucleotide polymorphisms (SNPs) markers that closely associated with chalkbrood-resistance and used the markers to make molecular assistant breeding. One SNP located in the second intron of MRJP5, termed as SNP-3, was closely related to chalkbrood-resistance and verified though rigorous experiments. The frequency of C allele (PC) of SNP-3 was significantly higher in chalkbrood-resistant colonies than that in chalkbrood-susceptible ($P < 0.05$). Another SNP was screened and verified within gene LOC100578413 named SNP-11 located in noncoding region of chromosome 11. The SNP-11 was associated with larval resistance to chalkbrood disease and the T allele frequency of this SNP in resistant larvae was significantly higher than susceptible larvae.
The chalkbrood resistant honey bee line showed certain resistant level to chalkbrood by the comparison of SNP-3 among different larvae samples. SNP-3 was further supported to identify the resistant level of the colonies using the PC. The colonies headed by C/C queens might have more resistance to chalkbrood infection. These results suggested that SNP-3 could serve as the genetic marker to breed resistant colonies and queens conferred by resistant larvae themselves.
These results provided one convenient molecular marker for selecting honey bee chalkbrood resistant line Fengqian No.1 Italian honeybee, which was helpful to reduce impacts of chalkbrood on apiculture.

Quantification of the sensitivity and communication responses of high and low-grooming honey bees (*Apis mellifera*)

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The invasive ectoparasitic mite Varroa destructor is a main driver of honey bee colony losses around the world. Currently, chemical miticides are the most common controls, however, they are imperfect as they can lead to acaricide-resistant mites as well as chemical residues in wax and honey. Thus, capitalizing on natural social-immunity behaviours, such as grooming behaviour, to breed bees resistant to varroa mites may be a good alternative method of control and would help to decrease reliance on chemicals once resistance is established. Though much is already known about honey bee grooming behaviour, we sought to answer the question of whether bees coming from colonies exhibiting higher levels of grooming are more sensitive to stimuli than those coming from low-grooming colonies. In laboratory petri dish assays, we found that high-grooming bees spent significantly more time auto-grooming after receiving a stimulus of either a single varroa mite or a small puff of chalk dust applied to the thorax, relative to low-grooming bees. Further, when grooming responses were subdivided into light or intense, we found that high-grooming bees spent significantly more time intensely grooming when receiving a stimulus treatment of varroa or chalk dust applied to the head or thorax, relative to control bees receiving no stimulus. The results for a second experiment are currently being processed, where grooming behaviour at the colony level was examined using small cohorts of bees placed into experimental cages. Cages were again stimulated with varroa mites or chalk dust and the responses to be measured include allo-grooming, grooming invitation dances, sound, and heat using an infrared camera. This research will elucidate the specific mechanisms through which bees achieve enhanced grooming success against varroa and communicate among nestmates. This should then facilitate quicker screening methods for grooming behaviour and reveal novel targets for selection of proteomic and molecular markers that correlate with these traits, thereby speeding up the process by which high-grooming bees can be selected for in breeding programs.

BEE HEALTH

12 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

CONTROL OF BEE DISEASES AND PESTS I ROOM 517C

[Lead-off] The future of pest and disease control

E. Genersch
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The Western honey bee *Apis mellifera* is a generalist pollinator for numerous wild flowers and, therefore, it contributes to biodiversity in natural ecosystems. In addition, managed colonies of *A. mellifera* are widely used as commercial pollinators of many crops and fruit in global agriculture thus contributing to both the profitability of apiculture and agriculture. Hence, infectious diseases threatening the performance and survival of individual honey bees and honey bee colonies are of great concern for beekeepers and farmers alike. Among the pathogens posing the most serious threats to honey bees are the RNA virus DWV (deformed wing virus) vectored by the metazoan parasite Varroa destructor, the bacterium *Paenibacillus larvae* causing the notifiable epizootic American Foulbrood (AFB) and the microsporidium Nosema spp. as causative agent of dysentery. Recent discoveries and developments have considerably improved our knowledge on these pathogens. We now understand much better how the Varroa mite and DWV interact which might help to combat both. A recently developed cell culture model for Nosema spp. already aided in identifying novel substances active against this obligate intracellular parasite. And our detailed knowledge about virulence factors differentially expressed by the most prevalent P. larvae genotypes ERIC I and ERIC II might pave the way for developing sustainable measures against AFB. These are all examples showing how research and development shape the future of pest and disease control.

Apiguard® efficacy for controlling Varroa destructor in honey bee (*Apis mellifera*) colonies in Canada

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Apiguard® is a varroacide with thymol incorporated in a gel matrix. Apiguard® has two complementary modes of action: 1) bees ventilate and thymol vapours spread within the colony; 2) bees transport and spread the gel in the colony by trophallaxis. The thymol then eliminates varroa mites by contact. When Apiguard® is used following the label instructions, it is non-toxic for humans and well tolerated by bees. The aim of this work is to conduct an efficacy trial of Apiguard® under typical Canadian apicultural climates and management practices. We tested three different treatments on groups of 12 colonies: 1) negative control (no treatment), 2) Apiguard® dosage 100g/colony/6 weeks (2 consecutive applications of 50 g), and 3) Apiguard® dosage 75g/colony/6 weeks (3 consecutive applications of 25 g). Treatments were compared using varroa drop during the treatment period and by calculating efficacy after a follow-up treatment with Apivar® (active ingredient: amitraz). The highest efficacy measured was with the Apiguard® dosage of 2 consecutive applications of 50 g (group 2; 89.8% ± 0.8, $p < .0001$). Furthermore, our results show that this Apiguard® dosage is well adapted for the Canadian climate because environmental temperatures are within the recommended temperatures by company. Temperatures decrease rapidly from September to October and applying a third dosage (as group 3) would not be efficient against varroa.

Laboratory, semi-field and field trials to assess the efficacy of Bacillus thuringiensis formulations against the larvae of the greater Wax Moth (*Galleria mellonella*)

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Wax moth larvae (WML) can destroy honeybee wax comb and cause serious economic losses for beekeepers. In recent decades, registered products based on Bacillus thuringiensis (BT), such as B401, have been applied to protect combs. In a two-year research programme, we tested BT products for their efficacy against WML and their safety for honeybees under laboratory, semi-field and field conditions.
A starter culture of WML was used for laboratory and semi-field trials, and early instar (7-day-old) larvae, weighing 0.2mg, were used. For the laboratory tests, one larva was placed into each well of a 24-well plastic tissue culture plate containing 0.25g of artificial diet.
Two formulations of BT, Bta and Btk (produced from different BT subspecies) were diluted in water and tested at three different doses. The controls were water and B401 (positive control). A total of 48 replicates was used for each BT treatment (96 for the water control), each replicate consisting of one larva per batch, giving a total of 864 replicates. Each product was applied via the moths' artificial diet. After incubation, dead larvae were scored at days 4, 7 and 11. The most effective application was Bta (rate 1:35) followed by B401 (efficacy: 95.83% and 75%, respectively). When compared with the controls, Btk presented no significant difference. All applications limited the ability of surviving WML to spin cocoons. Control larvae cocoon spinning was unaffected.
During semi-field trials, the products were tested on 10x10 wax comb, with 20 WML isolated in plastic containers on each comb. Both

WML mortality and the combs' undamaged area were measured. Application of Bta at rates of 1:17.6 was the most effective, inducing 99.66% mortality on WML and protecting 99.13% of the comb area.
In field trials, all products appeared to be safe for honeybees during tolerance tests in colonies equipped with Gary traps. The number of dead bees presented no significant differences to control colonies for all products tested.

Varroa treatment of brood-free honey bee colonies with lithium chloride

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Lithium chloride (LiCl) represents the first new and promising acaricidal compound for the treatment of varroosis since more than 25 years. The systemic mode of action allows an "easy to apply" treatment via feeding. In cage experiments we could confirm a nearly 100% efficacy and a good tolerability of adult honey bees. However, the low lithium tolerability of larvae has so far limited the application in breeding colonies. We therefore performed first field experiments with brood-free colonies. In the first experiment, we established 34 artificial swarms and fed them for three days with either syrup only or syrup containing lithium salts at different concentrations up to 50mM LiCl. The swarms were then installed and a final treatment with Bayvarol® was performed. In the second field test, we created a brood-free period by caging the queens of 9 honey bee colonies for 21 days. The colonies then received either syrup with 25mM LiCl or sugar dough with 50mM LiCl over a period of 5 days. Four additional control colonies were treated once with formic acid. Final control treatments of all colonies were performed with Bayvarol®. In both experiments, the short-term applications of lithium salts revealed efficacies ranging from 82% to 98%, while visible brood damages could not be observed. Further, the lithium treatment revealed a significant higher efficacy compared to the recommended formic acid treatment. However, a crucial factor for the efficacy of a lithium treatment is a rapid distribution of LiCl among all bees of a colony. We therefore colored both, syrup and dough with food dyes (Ehrenberg et al., 2019) and analyzed worker bees from different parts of the hive at the end of the LiCl treatment for coloration of the digestive tract. Surprisingly, more than 91% of the bees had contact with the colored food – independent of task and position in the hive. This indicates that even with a short-term treatment of LiCl a high efficacy in full-sized colonies is possible. However, the crucial challenge is still the registration of LiCl as a veterinary product that requires the support of a competent company.

Impacts of requeening practices on honey bee health

A. Dalmon [1], V. Desclos [1], C. Kouchner [1,2,3,4], M. Pioz [1,2], B. Basso [2,3], M. Ventelon [1,2], J.L. Brunet [1,2], A. Decourtye [2,3], Y. Le Conte [1,2]
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More than 30 viruses infecting *Apis mellifera* have been described. Among them, five viruses are widely spread in the colonies all around the world, and the Deformed wing virus and the Sacbrood virus have been shown to contribute to colony losses. Most of them are transmitted by the vector Varroa destructor, but they can be transmitted both horizontally and vertically, i.e. from the queen and/or drones to their progeny. Thus, the queen management practices and especially the requeening practices of the beekeepers may impact the virus loads in the colonies.
In order to assess the impacts of an annual requeening compared to natural supersedure, we monitored a three-year experiment comparing these two strategies. Within each of these two strategies, two honeybee strains are compared in two different apiaries (120 colonies in total): a "requeening strain" from a beekeeper who annually requeens his colonies, and a "natural replacement strain" from a beekeeper who lets his colonies naturally replace their queens. Thus, this experiment allows to consider the effects of both queen management practices and honeybee strain on the health status of the colonies.
Since the beginning of the experiment in 2016, the viral loads of the colonies were followed annually in spring and autumn by quantitative PCR and varroa pressure was assessed at the end of each summer, after the honey harvest. Through this experiment, we investigate the possible impacts of two contrasted queen management strategies on the evolution of the colony viral loads over three years. The possible consequences of requeening on the colony health status are also considered through the comparison of the queen viral load to the mean viral load of the colony workers.

The small hive beetle world tour: role of solitary bees, flowers and wax

P. Neumann, F. Ouessou Idrissou, Q. Huang, O. Yanez
Institute of Bee Health, Vetsuisse Faculty, University of Bern, Bern, SWITZERLAND

Small hive beetles (SHBs) are parasites of social bee colonies endemic to sub-Saharan Africa. They have become an invasive species and have now established populations on all habitable except Antarctica. Outside of their native range they can cause considerable damage to apiculture and may also endanger wild bees, thereby creating demand for better mitigation. The talk will give an overview on recent research, including novel evidence for the global spread via apicultural trade, the role of solitary communal nesting bees as alternative hosts as well as the previously overlooked role of flowers for SHB survival. In summary, enhanced border control for trade of beehive products combined with improved control, taking into account the role of wild bee reservoirs and flowers, appears prudent to limit the future spread and impact of this invasive species.

Mating and survival of small hive beetle Aethina tumida

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Mating behaviour and adult survival in response to starvation are two important parameters connected to the success of any animal species. A lack of respective knowledge often hampers mitigation efforts for invasive species. This holds especially true for small hive beetles (SHBs), Aethina tumida Murray (Coleoptera: Nitidulidae), which are parasites of bee colonies endemic to sub-Saharan Africa. In their new range, they can cause serious damage to colonies of European honeybee subspecies. Even though SHBs are sexually reproducing animals, comparatively little is known about the actual mating behavior of this species. Moreover, the survival of adult SHBs under pressure from starvation is currently unknown, even though this appears to be crucial for estimating invasion risk under various pathways. Here, we studied SHB mating behaviour and survival in a laboratory experiment that manipulated the beetle diet. The results of the ongoing experiments will be presented and discussed.

SYMPOSIUM 13:00-15:00
CONTROL OF BEE DISEASES AND PESTS II ROOM 517C

Wintering Method and Nosema Control in Honey Bees (*Apis mellifera*) under Canadian Prairie Conditions

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Infection of honey bee colonies with Nosema spp. has been reported to reduce adult bee populations and brood production, leading to increased winter colony losses. Alberta, one of the Canadian Prairie Provinces, experiences long, cold winters, and Nosema is typically among the top four factors causing colony mortality. Climatic differences across the province, as well as differences between wintering method (indoor vs. outdoor), can influence Nosema mean abundance in colonies. Fumagillin treatment, which has been shown to reduce Nosema abundance, can be applied in the spring or fall, but a need for both seasonal treatments has not been shown. The objective of this study is to determine the effect of geographical location, wintering method, and fumagillin treatment timing on Nosema abundance and honey bee colony strength and winter survival in Alberta. From May 2017-18, a total of 128 colonies were monitored which were split equally between Edmonton (Northern Alberta) and Brooks (Southern Alberta). In each location, colonies were randomly assigned to one of the following fumagillin treatments: spring only, fall only, spring and fall, and non-treatment control. For the winter, colonies in each location are split equally between being wintered indoors and outdoors. A hemocytometer and quantitative real-time polymerase chain reaction (qPCR) were used to determine spore load and species, respectively. Nosema abundance was affected by both location and wintering method. Fumagillin treatment reduced Nosema abundance in colonies, however, the treatment did not significantly increase brood area or adult bee population in the months leading up to winter. Though, brood area and adult bee population were significantly greater in Northern than Southern Alberta. Colony winter mortality rates were affected by both wintering method and location. This research provides beekeepers with much-needed information to make appropriate integrated pest management decisions to combat losses due to Nosema.

Effect of selected prebiotics and probiotics on the parasitic fungus Nosema ceranae and on the health of honey bee colonies

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Nosema ceranae and N. apis are fungal parasites of the honey bee ventriculus that cause nosema disease. For over 60 years, fumagillin has been the only antibiotic used to treat Nosema spp. in honey bees. While effective, fumagillin poses concerns of contamination of hive products with residues and the commercial formulation of fumagillin sold in North America has been recently discontinued. Clearly, alternative compounds are needed to control Nosema infections in honey bee colonies. Several natural compounds, including prebiotics such as eugenol (active component of clove oil), chitosan (chitin from the shells of crustaceans), and naringenin (flavone from citrus fruits), as well as one probiotic (a commercial formulation of Enterococcus faecium), were evaluated in field honey bee colonies for their effects on N. ceranae infections and bee health parameters. The compounds were administered in solid (protein patty) or liquid (sucrose syrup) form to the colonies. Eugenol, naringenin and the E. faecium probiotic, reduced N. ceranae infections and increased honey production, while the probiotic also increased adult bee populations and eliminated colony winter mortality. Chitosan was ineffective. In general, compounds administered in protein patty worked better than in sugar syrup in spring, whereas sugar syrup worked better than patty in fall. For colonies, the commercial formulation of E. faecium was the most promising candidate for controlling N. ceranae and promoting honey bee health and productivity, warranting further investigation.

Development of a novel point-of-care diagnostic test for foulbrood diseases of honey bees

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American and European foulbrood (AFB & EFB) are devastating bacterial brood diseases of *Apis mellifera*, which cause colony and economic losses worldwide. Both diseases show a patchy brood pattern often with sunken cell cappings, a specific odor and infected larvae may lose their tissue integrity. In the field, trained persons perform a visual diagnosis of the suspected colony brood nests to distinguish between both diseases. However, in the case of AFB, two ERIC-genotypes that differ in virulence cannot be distinguished by visual diagnosis. Therefore, a reliable, specific and sensitive tool to detect the causative agents of AFB and EFB (*Paenibacillus larvae* & Melissococcus plutonius) and to distinguish between genotypes ERIC I and ERIC II of P. larvae would be of great help to allow a fast field diagnosis to decide how to proceed in control of the diseases.
The aim of the project is the development of a fast and sensitive field test kit (lateral flow device) to diagnose and distinguish between EFB, AFB and two AFB-genotypes (ERIC I & ERIC II) in one device. Pathogen-specific antibodies are generated by characterizing the humoral immune response to the three different causative agents in mice, rats and rabbits. The sensitivity and specificity of the monoclonal antibodies (mABs) were determined using ELISA & Western blot analysis. Specifically detected antigens of the different pathogens will be identified and characterized using mass spectrometry. Applying optimized mABs and purified antigens, a highly sensitive lateral flow device based on multicolored silver Nano plates for multiplexed diagnostic will be established. Finally the novel point-of-care test will be validated (specificity and sensitivity) using field samples from disease outbreaks.

Lessons learned from developing an RNAi-based varroa control product

J. Masucci, D. Avni, A. Inberg, N. Durnell, S. Back
Bayer Crop Sciences, St. Louis, USA

Beekeepers and bee researchers agree that Varroa mites and the diseases they spread are the top threat to honey bee colonies today. The combination of Varroa infestations with other stress factors bees encounter results in high losses of honey bee colonies annually. Several mite treatments are available on the market, but they target phoretic mites and fail to impact reproducing mites. Due to environmental factors and mite resistance many treatments are not appropriate in some regions resulting in the over-use of a single product. Over the past several years, we've been conducting large field trials to develop a novel, RNAi-based mite control product called BioDirect™. Because RNAi is so specific, there is no detrimental effect on the bees. Therefore, BioDirect™ is applied in sugar syrup and uses colony dynamics to ensure its delivery directly into the brood cells by the bees. Varroa mites are exposed to BioDirect™ when entering the brood cells to reproduce, thus providing a time of action complementary to commercially available treatments. The field trials themselves are some of the largest honey bee field trials conducted and what we've learned from these trials is useful to beekeepers and researchers alike. The large data sets informed us on mite population dynamics, mite control, and the impact on honey bee health. We will report how trial design and mite migration can influence the outcome of the experiments and show how mite populations recover from a commercially available treatment. Our data show that BioDirect™ reduces mite infestation levels and results in colony survival rates similar to the commercial standard. BioDirect™ may provide a novel mode of action to be used as part of an Insect Resistance Management system for controlling Varroa mites in honey bee colonies.

Climex modeling for spatial expansion of its distribution, Vespa velutina, a predatory hornet after the invasion into Korea: Model development and validation

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Vespa velutina nitrothorax was introduced into Korea in 2003 and then with some lag period, its spatial distribution has been in expansion. For the modeling of the spatial expansion, we did the spatial modeling based on the climatic conditions and biological adaptive capacities using Climex modeling tool. Model estimated that the climate changes with warming positively affected the spread of the invaded, south-origin species. Also based on the scenario, RCP 8.5, southern part of Korea would be highly relevant for the habitat suitability for the invaded Vesap establishment. In this study, we used ecophysiological parameters and meteorological data in CLIMEX to calculate an annual growth index (GIA). This index helps to understand the potential for population growth of the invaded species during favorable conditions, and up to eight stress indices (heat, cold, wet, dry stresses and their interactions) to simulate its ability to survive unfavorable conditions. Further nation-wide monitoring were performed in the fall season as well as spring season. Total of 136 county-cities were encompassed for the study out of 240 city-counties nationwide. The results clearly showed that the invaded species, Vespa velutina from the Southeastern tip of the peninsula is now distributed across the south part of Korea including some remote counties. Higher densities were noted on the lower latitudinal sampling sites. Density-gradients were somewhat independent to its main prey species availability,

density of hive of honeybees. These clearly demonstrated that the temperature would be one of the most important factors for its survival and distribution. Further discussion were made on the suppression of the invaded vesap for the protection of honeybee and other non-target effects.

Using of Jatropha oil extract for controlling Varroa mite in honeybee colonies

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Varroa destructor is a severe external parasite mite of the honeybees and caused heavy losses of colonies globally. Present work evaluated the effectiveness of Jatropha oil against Varroa infested honey bee colonies. Four concentrations, 1%, 2%, 5%, 10% of Jatropha curcas oil were prepared for controlling colonies infested by Varroa. Twenty honey bee colonies of Carniolan hybrid *Apis mellifera L.* naturally heavily infested by Varroa mites were used for experimental procedure. There were significant effects of reducing percentage of Varroa mite infestation on sealed brood and adult workers (P= 0.05) after 2nd, 3rd and 4th treatments for sealed brood and after 3rd and 4th treatments for adult workers in all tested concentrations of Jatropha oil. The low concentrations 1% and 2% of Jatropha oil had the heist reduction of percentage of Varroa infestation, while the high concentrations 5% and 10% had the lowest reduction of percentage of Varroa infestation. Moreover, the colonies treated with the lowest concentrations of Jatropha oil had the highest amount of brood area (75.75 and 77.50 inch2) and the highest number of combs covered with bees compared with the colonies treated with concentrations 5% and 10% of Jatropha oil. Treated colonies with concentrations 1%, 10% and 5% had a high amount of stored honey and pollen grains (126.50, 111, 96 inch2) and (11.25, 9.75 and 9.75 inch2), respectively.

Development of detection method as point-of-care using Ultra-rapid PCR and immunochromatography against 11 major pathogens in honeybee

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We have developed a detection method as the point-of-care using ultra-rapid PCR and Immunochromatography against 11 major pathogens of honeybee. The 11 kinds of major infectious pathogens in honeybee included Black queen cell virus (BQCV), Chronic bee paralysis virus (CBPV), Deformed wing virus (DWV), Israeli acute paralysis virus (IAPV), Sacbrood virus (SBV), Korean sacbrood virus (kSBV) as infectious viruses, *Paenibacillus larvae* causing American Foulbrood (AFB), Melissococcus plutonius causing European Foulbrood (EFB) as bacteria, Ascosphaera apis (Chalkbrood), Aspergillus flavus (Stonebrood), Nosema apis and Nosema ceranae (Nosemosis) as fungi. The specific primer pair of each pathogen was designed for ultra-rapid PCR detection. The developed PCR-chip-based ultra-rapid PCR showed successful amplification for all 11 major pathogens in the presence of more than 102 molecules. The time required to confirm the amplification (Ct-time) was about 10 minutes. Total PCR analysis time, including PCR for a total of 50 cycles, melting point analysis and reverse transcription, was approximately 20 minutes. This detection method is capable of rapid and high sensitivity testing by combining ultra-rapid PCR and Immunochromatography.

Data-driven educational programming empowers beekeepers to effectively manage colony health

E. Mullen
Cornell University, Ithaca, USA

Varroa mites are the leading cause of colony losses in most countries, including the United States. Cumulative research over the past decade has yielded substantial insights into the impact of Varroa on colony health and has guided the development of best management practices to control this parasite. Translating the latest scientific research to the beekeeping industry and providing training to beekeepers that equips them with the skills to effectively manage Varroa is critically important to improve colony health and reduce colony losses on a large scale. Yet few training programs of this nature exist, and the impact of such training programs are rarely known since longitudinal data regarding their success are rarely gathered. In this talk, I will show evidence that the NYS Beekeeper Tech Team, an educational program that translates science into practice, successfully empowers beekeepers to effectively manage Varroa and other colony health metrics by adopting evidenced-based practices.
The New York State Beekeeper Tech Team works with beekeeping operations to improve colony health by providing information and training to beekeepers. Participants regularly receive field visits, parasite data from their own colonies, and training from an interdisciplinary group of researchers and extension personnel to strengthen their skills in managing Varroa using an Integrated Pest Management approach. Survey data are paired with colony sampling data to evaluate changes in colony health and management over time. This data-driven educational program has led to substantial improvements in Varroa management by beekeepers and, as a result, improved colony health.

Within three years since the program's inception, beekeepers have experienced fewer colony losses, monitored for and treated Varroa more often, had fewer colonies with Parasitic Mite Syndrome, and had fewer colonies that were above the treatment threshold for Varroa mites. This program can serve as a model for other organizations and academic institutions that assist their regional beekeeping industries, and the results shared in this presentation can also provide insights to additional beekeepers in developing their own Varroa management plans.

Developing alternative new synthetic miticides for Varroa mites control

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Plant And Bee Health Surveillance Section, Alberta Agriculture And Forestry, Edmonton, CANADA

One of the greatest threats to the honey bee worldwide is Varroa destructor. Given the complex interactions between the honey bee and Varroa mites, synthetic miticides play a major role in the management of Varroa. Within a few years of repeated use of the same miticide, beekeepers began to report the development of resistance to applied miticide. Our research objectives are focused on screening miticides with different modes of action to provide effective options for mite control and management of resistance. Bioassays were established for screening 22 active ingredients (AIs) and their formulated products (FPs) belonging to different family classes of miticides, for their efficacy on Varroa and safety to bees under laboratory and field conditions during 2016-2018. In this study, the positive control was the common miticide, Amitraz, with no treatment as the control. To determine LC50 of tested products for the mite and bee mortalities, 20 ml glass vials and 60 ml Masson jars were used for mites and bees, respectively. Each vial or jar was treated with 0.5 ml of each serial dilution of tested compounds. In each vial, 8-10 mites were used and in each jar 10 bees were used. The LC50 was then calculated after 24 h. To determine LD50 after 24 h, a group of 10 mites and 10 bees were topically treated with 0.15 µl /mite and 1 µl/bee of each serial dilution of tested compounds, then placed in vials and jars, respectively. Results indicated the eight of the tested AIs were potentially effective against mites and safe for bees. FPs of promising AIs tested in the lab were evaluated in the field, with Apivar used as positive control. Single brood chamber colonies constructed with three separate compartments, three combs each. Varroa infested bees covered three frames were used. Mite mortalities were determined using sticky boards and efficacy calculated based on using Apivar as a finishing treatment. The field results identified two potential acaricides groups, Pyrazole and Quinazoline, both effective against mite and safe for bees. This is an important first step to developing new synthetic miticides for use in Varroa management systems.

POLLINATION AND BEE FLORA

09 SEPTEMBER 2019

SYMPOSIUM 15:30-17:30

STATUS & CONSERVATION OF POLLINATORS ROOM 517D

[Lead-off] Pollinators and pollination under pressure: problems and progress with this predicament

N. Raine

University of Guelph, Guelph, CANADA

Roughly one third of the food we eat depends on the services of pollinators. Economically sustainable yields for 75% of global crop types are byproducts of pollinator foraging activity (most importantly bees), moving between flowers collecting food and also carrying pollen to facilitate seed, fruit and nut production. Beyond crops, almost 90% of flowering plant species worldwide rely on animal-vectored pollination, making pollinators an essential part of natural ecosystem function and wider cultural values. Reports of extensive and widespread global pollinator declines raise concerns for agricultural productivity, food security and reduced natural biodiversity. Declines seem to be driven by multiple, potentially interacting environmental stress factors. These include the loss and fragmentation of habitat, increased agrochemical exposure resulting from agricultural intensification, impacts of parasites and pathogens, invasive species and climate change. In this presentation I will review the evidence for, and impacts of, pollinator declines and discuss potential strategies to enhance pollinator health and sustainable agricultural production. The focus and scope of management or mitigation strategies might differ substantially depending on desired conservation outcomes. For example, supporting the habitat requirements of key pollinators (species or communities) for sustainable production of a particular crop might be appreciably different to the needs of targeted pollinator species assessed to be at risk of extinction. Pollinators are beautiful, fascinating, diverse and essential creatures that we simply cannot afford to lose.

Impact of agricultural intensity on bumble bee communities in Southern Québec: a 10-year story

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Bumble bees, as many other insects, are in decline worldwide. Many factors have been put forward to explain such declines, including parasites, climate changes, and particularly, agricultural intensification with its monocropping systems, loss of natural habitats and its significant use of pesticides. Studies investigating the effect of agricultural intensity on pollinators at the landscape level are usually performed using relatively short-term (1-3 y) datasets. Here, we use a 11-year time series (2006-2016) of capture data from 40 farms located in southern Québec, Canada, in order to quantify the impact of agricultural intensity on the structure of bumble bee communities. We hypothesized that intensive cropping systems (e.g., corn, soybeans) would be the most important drivers explaining bumble bee community assemblages. We also hypothesized that the arrival of neonicotinoid-coated corn and soybean seeds in 2008 would result in a quick turnover of species. We tested these hypotheses based on bumble bee samples from yellow window traps (2 / farm) that were collected every 2 days from May to July. Bumble bees were identified to the species-level and queens, males and workers were sorted out. Landscape composition within 500 meters of traps was determined every year in situ and using data from ‘’La Financière agricole du Québec’’ for radii of 1, 5, and 10 km. We used a hierarchical modelling of species communities (HMSC) approach to assess which landscape components influenced bumble bee communities as well as to determine if these effects were associated to species-specific functional traits, such as body size and tongue length. Our study shall therefore shed light on the temporal response of bumble bee communities facing agricultural intensification, as well as on the combination(s) of phenotypic traits that put some species more at risk than others in such a context.

Pollinators as indicators in policy affecting the landscape and environment

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Bees live in very close contact with their surroundings, from which they get not only food and water sources, but also materials and/or habitat to build their nests and reproduce. Furthermore, bees and honeybees in particular, are the link between nature and culture. They are a key agricultural agent which can provide essential insights to assess results of land management practices. Both wild and managed bees, as well as many other animals, are crucial for our food security due to their pollination activity. A decrease in their abundance and diversity are putting at stake our way of life and the range of the food offer. Civil society is increasingly demanding public authorities to establish more transparent forms of accountability. For this purpose, we preconise here a Pollinator Index as a proxy indicator for: (1) pollination services; (2) environment quality and health; (3) result measurements in terms of sustainability of policies implemented in agricultural or land management. Such an index would be a tool to monitor the real performance of different public policies and their impact on the environment, which could allow the calibration of public spending towards improving measures/policies that target pollinators. Bearing in mind the differences between the biology and habitats of different pollinators, we envisage that such an index would be composed of different elements, namely: (a) rate of winter/summer honeybee colony losses; (b) wild bees abundance and richness; (c) honeybee collected pollen pellets botanical origin and contaminant content; (d) amount of honeybee colonies per km2. By verifying the evolution of such an index, as an "impact indicator", decision makers could establish goals and strategies, and even point towards the tactics to achieve them. Furthermore, a Pollinator index would contribute to increase transparency in policy results, providing citizens a tool to verify if public money is invested for the preservation of public goods.

The Oregon Bee Project: Working together for pollinator protection in Oregon, USA

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Oregon has one of the most diverse, vibrant, and dynamic group of bees in the US. This not only includes 80,000 commercial honey bees colonies, but three other managed species and at least 500 wild species. This level of species richness is supported by the diversity of both natural and managed habitats found within the state, including the more than 200 crops grown in Oregon. However, declines in honey bee populations, concerns about wild bee populations, and a series of high-profile bee kills from the misuse of pesticides have catalyzed efforts to protect and improve pollinator health. The formation of the Oregon Bee Project in 2018 was a response to these concerns, state legislation and federal directives. Many states in the US that have developed Managed Pollinator Protection Plans (MP3) documents, stemming from requirements from the US Environmental Protection Agency. The Oregon Bee Project, however, has operationalized its plan through an array of educational, outreach and decision-support services delivered through cooperative Extension, state agencies and volunteer programs (e.g., Master Beekeepers and Master Gardeners). The paper discusses how the Oregon Bee Project was initiated, its specific goals and assessment metrics and the challenges associated with MP3 process in the US.

Forest proximity maintains Bumblebee species richness in a boreal agricultural landscape

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To create functioning conservation measures for pollinators, we need to understand how landscape structure affects their effectiveness. We examined the effects of forest proximity on *Bumblebee* communities in a modern agricultural landscape in the boreal region. We contrasted *Bumblebee* communities in field margins bordered by forest with margins located next to fields. We also measured the effect of forest proportion in the surrounding landscape on *Bumblebee* species richness. Additionally, the effects of margin area and flower abundance on *Bumblebee* species richness were assessed. As expected, species richness was higher in margins bordered by forest compared to margins next to fields. Higher proportion of forest in the surrounding landscape, as well as larger edge area and higher flower abundance, also resulted in higher species richness. Notably, these effects occurred only, or were considerably stronger, in margins next to forest. Our results show that forest patches have to be preserved in order to maintain or increase pollinator species richness in agricultural landscapes. Margins next to forest should be preferred while planning local conservation measures.

The Role of Insect Pollinators in Sustainable Agriculture: An Overlooked Agricultural Input in Ethiopia

T.K. Gemeda, K.W. Hora

Oromia Agricultural Research Institute, Holeta Bee Research Center, Holeta, ETHIOPIA

Sustainable agriculture is a function of natural ecosystems outcomes than specific agronomic practices. Pollination service is agricultural inputs that ensure the production of crops and smallholders around the world benefit from the services. The current expansion of pollination-dependent crops together with the declining scenario of pollinators and their habitat recently raise concerns of possible yield reduction in agriculture. Improving pollinators' diversity and density has direct positive impact on crop yields, consequently promoting food and nutrition security. Underestimation of the essential role played by managed and wild insect pollinators is key constraint to the sustainability of current agricultural practices. Insufficient knowledge among crop growers and crop pest control operators about the importance of pollinators and pollination services hinders the conservation and sustainable use of natural pollinators for sustainable agriculture. The intention of this review, therefore is to synthesize overviews on the role of pollination in sustainable agriculture, conservation strategies that could help to enhance pollinator habitats, the global perspectives of insect pollination, and to argue the current pollination situation in Ethiopia. Among the most critical priorities to ensure sustainable use of pollinators including (1) documentation of pollinator-dependent crops, (2) quantification of pollination gaps, (3) development of conservation mechanisms that enhance synergies between pollinators and crop production. Strategic coordination among crop growers, beekeepers, agricultural researchers, conservationist and policy makers to develop joint efforts to enhance local solutions for pollinators conservation, and ultimately agricultural productivity is very crucial. Moreover, capacity building through networking among farmers, extension workers and researchers is important to recognize the economic and ecological value of pollinators.

Conserving endangered indigenous, *Apis cerana* in traditional hives for sustainable beekeeping in Jammu and Kashmir

D. Sharma, D.P. Abrol, H. Ahmad
Skuast Jammu, Jammu, INDIA

Indigenous honeybee, *Apis cerana* have been playing a key role in by enhancing agricultural productivity as they are a natural pollinators for a range of fruits and crops. The *Apis cerana* is very popular among the farmers in rural areas, probably due to the lowest cost of construction of traditional hives (log and wall) via using locally available materials. There are around 12,662 colonies of *Apis cerana* in traditional hives, out of which 5,150 colonies of *Apis cerana* are kept in log hives and 7,512 colonies in wall hives. In Jammu region, The wall hive beekeeping is practiced in Jammu, Kathua, Reasi, Udhampur, Ramban, Poonch, Rajouri and Samba districts of Jammu region. The log hive beekeeping in practised in Poonch, Rajouri, Doda and Kishtwar districts of Jammu region. The *A. cerana* strain of Jammu and Kashmir is not only larger but also higher honey-yielding than other counterparts. Unfortunately, *A. cerana* is threatened in Jammu and Kashmir as beekeepers are confronted by development extensionists trying to encourage introduction of *Apis mellifera* – in the areas of origin of *Apis cerana*. Integration of the traditional method with the modern concept of movable frames would add to the ease of management operations in the field making the modernized wall hive an eco-friendly, readily acceptable, economically viable and environmentally sustainable technique for the future. There is an urgent need for organizing village-based training and building the capacity of local beekeepers as trainers to provide follow-up support. More research is needed to improve the indigenous methods and adapting to modern scientific methods of bee keeping. Creation of research projects at local level focussing with diversity, conservation and sustainable beekeeping. Traditional beekeeping with *A. cerana* should be encouraged among rural households for increase crop pollination and sustainable development.

POLLINATION AND BEE FLORA

10 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

HONEY BEE POLLINATION OF CROPS ROOM 517B

[Lead-off] Training honey bees to commercial crops

W.M. Farina

Social Insects Lab, Department of Biodiversity and Experimental Biology, Institute of Physiology, Molecular Biology, Buenos Aires, ARGENTINA

The main economic contribution of the honey bee Apis mellifera is not the honey and other products obtained from their hives, but their ability to pollinate the most varied agricultural crops, including fruits of high quality and seeds for oils. As an established insect species worldwide, its use as a pollen vector generates significant profits in terms of agricultural production. Agronomic and veterinary sciences have made notable contributions to improve the relationship between honey bees and crops. However, knowledge from the biological sciences, such as honey bee behavior, physiology and ecology have been less taken into account in order to achieve an improvement in pollination efficiency and crop yield. Nowadays it is clear that the sophisticated honey bee communication systems together with its cognitive abilities are nodal to understand how individual and social information is used during the collective search for resources. With the aim of improving the bee-crop relationship, our laboratory has developed an interdisciplinary project focused on the cognitive ecology of the honey bee in commercial crops. Within this frame, a platform of environmentally friendly formulations promoting the directed pollination of these insects in crops has been developed. These tools not only increase crop yields, but also improve the sanitary and population status of colonies used in pollination services. On this talk, I will focus on some results that we obtained in different commercial crops.

Evaluation of the pollination deficiency in cranberry crops with the Stevens variety and a honey bee hives stocking rate of 2 hives/ac

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Although some cranberry growers use more than 4 hives/acre, the current recommendation for colony density in Quebec is 2 hives/acre and there are very few scientific publications to support this recommendation. For 2 years, on five farms using a stocking rate of 2 hives/ac, 3 fields of the Stevens cultivar distanced of about 750 m were selected and lots of 2, 4 or 8 hives were placed during bloom. At 50 m of those colonies, 5 plots were identified in a transect and for each, 20 stems with a potential of 4 fruits or more were selected (20 stems x 5 plots x 3 fields x 5 farms). Fruit set and fruit weight were monitored on those stems. The normal hives distributions on the farms were in lots of 40 to 120 colonies. Our results showed an increase in fruit set and fruit weight for the lots of 8 hives vs 2 hives. This would translate to a yield increase between 13 and 18%. Because of the hives distribution pattern on the farms, this yield increase can't be generalized for the entire farm. New studies using different stocking rates on half the farm would provide more insight. We also found that the first 4 flowers account for 95% of the yield and the first 2 for 65%, this indicate that pollinators management could be change for pesticides application in late bloom without compromising yields.

Pollination by honeybees enhances apple yield and fruit quality in Chitral, Pakistan

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Apple is an important cash crop and plays a crucial role in the livelihoods of farming community in Chitral, Pakistan. The commercial varieties of apple planted in orchards in Chitral, for example Kala Kola (Red Delicious), Choupush (Royal Delicious), Royal Gala, are reported to be partially or completely self-incompatible and require cross-pollination for commercial yield and fruit quality. Though honeybees are reported to play an important role in apple pollination, there is little awareness about the role of honeybees among farmers, development workers, as well as at policy level in mountain areas including Chitral.

ICIMOD and AKRSP Pakistan, therefore, conducted the action research by engaging 27 apple growers in six villages representing different altitudinal locations in Chitral for two consecutive years (2014 and 2015) to demonstrate the benefits of using honeybees for apple pollination and its impact on fruit yield and quality to the apple growers, other farmers, extension workers in district horticulture/agriculture departments, and district level decision makers, and generate evidence on the role of honeybees as significant ecosystem service (pollination) providers to help establish mechanism on payment for ecosystem services.

The findings revealed that supplementary pollination by honeybees enhanced fruit set by 9.8% to 14.5%, and reduced premature fruit drop

by 1.6% to 6.4% and the overall fruit yield by 47.9% at different sites, besides, improving the fruit quality i.e. weight, shape and color of apple fruit.
Engaging farmers in this research helped raise awareness and enhance understanding about the importance of honeybees as pollination service providers. The results suggested that integrating pollination into the whole production and marketing system is important for bringing higher income and improved livelihoods – not only of apple farmers but also the beekeepers – the pollination service providers, suggesting possibility of establishing strong business partnerships between marginal beekeepers and large orchard owners for enhancing social capital and cohesion. The information generated underpin crop management and policy decisions focused on promoting insect pollination services as well as conserving / protecting pollinators so that effective crop production can be maintained in the face of ongoing environmental change.

Pollinator preference among agronomic types of crops: the case of *Apis mellifera* for melon (Cucumis melo) pollination

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A great number of varieties, types cultivars and hybrids of most cultivated plant species have been developed but the attractiveness of a crop species to its pollinators is commonly assumed to be similar among agronomic varieties. In melon (Cucumis melo L.), great difficulties in directing honeybees (*Apis mellifera L.*) to the pollination of some agronomic types suggest that bees can discriminate between them and have preference for visiting flowers of some melon types. We investigated these selective visits in hybrids of five melon types, Yellow (Goldex), Cantaloupe (Zelda), Piel de sapo (Ricura), Charentais (Banzai) and Galia (McLaren), recording bee visits and sampling nectar and odor of flowers from each melon type and flower gender (male and hermaphrodite). Results showed that foragers visited significantly ($p<0.05$) more flowers of Cantaloupe than all the other melon types and Amarelo and Piel de sapo were also significantly ($p<0.05$) more visited than Charentais and Galia. Also, hermaphrodite flowers were significantly ($p<0.05$) more visited than male flowers in all five melon types. We did not found significant differences in size, shape and color among flowers of the melon types, but they differed in the number, type and function of volatile organic compounds (VOCs) released, from 23 substances predominantly attractants in Cantaloupe down to three predominantly repellents in Charentais. We observed significant positive and negative correlations between the amount of D-Limonene and Benzaldehyde (bee attractant) and α-Pinene (bee repellent), respectively, to the number of bee visits to flowers particularly in the Cantaloupe type. Also, we found that melon nectar is rich in sugars, amino acids and flavonoids, and phenylalanine and tryptophan are predominant in hermaphrodite flowers and directly related to the greater number of visits received by hermaphrodite flowers. Charentais and Cantaloupe were richer in these amino acids, while Galia was the poorer. We concluded that flowers of different agronomic types are not always equally attractive to pollinators and, in the case of melon, VOCs are responsible for bee preference among flowers of different agronomic types while nectar composition answers for bee choice for hermaphrodite flowers, both cases with important implications to pollination.

Effect of Larvae number and sequential introduction on honey bee colonies for pear pollination

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Pear (Pyrus bretschnederi Rahd.) exhibits full self-incompatibility and requires cross-pollination for fruit set. Hand pollination is the main way for fruit set of pear production in north China. Honey bees are important potential pollinators to replace the manual pollination for pear pollination. However, pear bloom is unattractive to honey bees. In present study, we tested the effect of larvae number and sequential introduction of honey bee (*Apis mellifera*) colonies on pollen collecting at the principle pear variety "Suli" orchard. Results showed that the weight and diversity of pollen collected by honey bee were affected by the number of larvae and sequential introduction. With the number of larvae increasing, the weight of pollen collection raised significantly ($p<0.05$). However, the pollen diversity was not affected by increasing the number of larvae ($p>0.05$). Sequential introduction of colonies could promote honey bee to focus on foraging pear pollen. Nonetheless, the pollen weight collected by sequential introduction colonies was not significant influenced ($p>0.05$). In conclusion, to promote the honey bee for pear pollination, we suggested to increase the number of larvae plus the sequential introduction colonies to the orchard.

Apis mellifera an efficient pollination alternative for strawberry and bitter gourd crops grown in enclosures

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India witnessed explosive growth in cultivation under enclosures but lags poorly in yield and quality components primarily due to non-fulfilment of pollination needs of crops. In order to develop viable pollination alternatives in strawberry (cultivar Winter Down) and bitter Gourd, Momordica charantia (cultivar Pusa Do Mausami) grown under polyhouses, readily and cheaply available *Apis mellifera* pollination units were employed in six different modes of placement viz. i) regular placement of colonies inside throughout flowering, ii) replacement with new colony after 15 days, iii) colonies with dual entrance, one opening inside and other outside enclosure on alternate days, iv) placement of colonies inside and outside enclosure on alternate days, v) hand pollination and vi) without insect pollination. Colonies were regularly fed with sugar solution. Bitter gourd seedlings were transplanted during rainy season (3rd July 2017) under net house and open fields. Results vividly revealed necessity of bee pollination as only 77 g/plant yield was harvested in without pollination plots. Yield increased significantly in open pollination (30.94 kg, 172 g/plant) but was still higher in manual and bee pollinated plots. Highest fruit yields (623 g/plant) were recorded in plots where *A. mellifera* colony was regularly kept inside followed by placement of dual way entrance colonies (584 g/plant). Colony replacement after 15 days and alternate placement in and outside enclosures had lower yields of 469 and 368 g/plant, respectively. These variations were observed over 18 pickings. Strawberry was transplanted during winter season (27th October, 2017) in naturally ventilated polyhouse and open fields. Lowest yield (295 g/plant) was again recorded in plots devoid of bee pollination. Regular placement of bee colony inside gave maximum yield (620 g/plant). Placement of colonies with dual entrance and replacement after 15 days yielded equitable yields (578 and 575 g/plant). Colony strength and forager population dwindled significantly when colony was regularly placed inside enclosures. Significantly higher forager population was maintained in colonies with dual entrance and when replaced with new colonies after 15 days. Study provides strategies to maintain strong bee forager force to ensure maximum pollination, overcoming the major bottleneck in the polyhouse, net house and other enclosures.

Towards best farming practices in BC blueberries to ensure bee health and good pollination

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Blueberries are Canada's top fruit export valued at over $400 million annually. In British Columbia (BC), the province with the largest production of cultivated (highbush) blueberries, crop productivity is dependent on the pollination service of managed honey bees. Bee pollination provides growers with greater fruit set, and produces larger and better quality berries. Blueberry pollination provides beekeepers with the first pollination income of the season and therefore both growers and beekeepers benefit from honey bee pollination.

Twenty thousand acres of mature blueberries are now grown in BC, requiring a minimum of 40,000 colonies for pollination each spring, and demand is increasing each year as more immature plants come into production. However, in recent years some beekeepers have become reluctant to engage in pollinating blueberries after observing poor health in their colonies, with a suspected increase in the incidence of European foulbrood (EFB)-like disease, following blueberry pollination.

In 2018, we initiated a project to study honey bee health during and after blueberry pollination. We conducted a field experiment with five commercial co-operator beekeepers engaging in highbush blueberry pollination. We compared pollinating and non-pollinating colonies and evaluated the effect of protein supplementation on several aspects of colony health, including adult population, brood area, and presence of diseases, noting an increase in EFB disease associated with blueberry pollination. We will discuss our findings and potential management strategies to increase pollinator health and pollination efficacy.

SYMPOSIUM 13:00-15:00

NON-APIS BEE POLLINATION OF CROPS — ROOM 517B

[Lead-off] Policies for pollinator-friendly agricultural landscapes

L.A. Garibaldi

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Pollinator diversity is being lost at an alarming rate. One of the main causes of this loss is the land-use change caused by the expansion of conventional agriculture and livestock production. Management practices such as monocultures and the intensive use of agrochemicals reduce the number of species of plants, birds, bees and other taxonomic groups, and increase, at the same time, the relative abundance (dominance) of one or a few cultivated and wild (e.g., weed) species. Given that ~40% of the terrestrial surface is occupied by crop and livestock lands, it is critical to increase food production without destroying pollinator diversity. In addition to the value given by its ethical and spiritual dimensions, and the potential use of future generations, in this talk I will discuss the value of biodiversity for agriculture, using pollinators as a case of study. Paradoxically, conventional agriculture is reducing pollinator diversity, but this diversity is necessary for increasing productivity (and its temporal and spatial stability) of many crops. Several studies show that the loss of wild pollinator diversity cannot be replaced by a high abundance of a single pollinator species (dominance). Therefore, I will discuss actions that producers, consumers, politicians and scientists can take to recover diversity. For example, producers can implement management practices in- and outside the crop fields to increase floral and nesting resources, and therefore pollinator abundance and diversity. In addition, consumers can modify diets, reduce waste and produce food at small scales, among many other actions. One single strategy will not be enough to solve the dilemma of producing food and preserving biodiversity: multiple actions must be taken urgently from all the stakeholders

Wild bees decline with distance into large strawberry fields regardless of edge habitat

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Flowering crops provide ample floral resources for many pollinators, but the time and duration of bloom and a lack of nesting habitat can reduce the suitability of cropland for many wild bee species. The preservation of potential pollinator habitat on croplands in the form of hedgerows, wildflower strips, and natural and semi-natural areas can help maintain wild pollinator populations in agricultural landscapes. However, the effectiveness of these enhancements in promoting wild pollinator diversity, and their effect on the pollination of the focal crops has been less explored. Here, we compared wild bee abundance and species richness in strawberry crops bordered by hedgerows and in crops bordered by more natural land (forests). We conducted bee diversity surveys in six strawberry fields with hedgerow margins and six strawberry fields with forested margins; all fields were at least 200 m long. We also examined strawberry pollen deposition at regular intervals into the fields and the magnitude of pollinator export from the field margins towards the center of the crops. Strawberry is an ideal crop to investigate pollinator export as the rows are covered with straw, which reduces habitat for ground-nesting bees within the crop; thus, most wild pollinators would need to enter the crop from the margins. We found that crops bordered by forests were no more species-rich or abundant in wild bees than crops bordered by hedgerows. Regardless of edge habitat type, we found that wild bee abundance and richness declined significantly from the crop edge towards crop center, but honey bee abundance did not. Strawberry pollen deposition also did not decline with distance into the crop, suggesting that managed honey bees (*Apis mellifera L.*) were providing most of the pollination at the field scale. Although our previous work indicates that wild bees are more effective (yield-increasing) pollinators of strawberry, their limited foraging ranges and lack of suitable habitat within the crop suggests they cannot provide full pollination to commercial strawberry fields in our area, given typical field sizes.

Landscape and farm management effects on bee assemblage and pollination in guava crop (Psidium guajava L.) in the semiarid Caatinga, Sergipe, Brazil

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Psidium guajava L. (Myrtaceae) is a tropical crop economically important in Brazil which can produce fruits throughout the year in semiarid irrigated Caatinga. Pollinators can improve crop yield and quality despite of the self-compatibility, but pollination services are not directly considered in management decisions by farmers. In this study we investigate the effects of landscape structure and farm practices on bee assemblages and its consequences on fruit and seed set and fruit quality; we also evaluated the influence of rural well-being assets (social, cultural, human, economic, physical and environmental) on the adoption of practices of ecological intensification through a participatory approach. We surveyed 10 conventional and irrigated small farms located in the municipalities of Canindé de São Francisco and Poço Redondo in Sergipe, Brazil. From May to December 2017, 740 bees were recorded; comprising nine species and seven genera, being Apidae the predominant family. Bee community was dominated by high density and the generalist bees *Trigona* spinipes and *Apis mellifera* (91.2% of the individuals), while no meliponini bee has been found and solitary bees were scarce and infrequent. Pollinating bees improved fruit set in 7.5% by reducing abortion rate (27%) compared to self-pollination (35% abortion rate, n = 108) and seed set (327 ± 30). Additionally, fruit quality was better (diameter: 63 ± 6 mm, length: 82 ± 12 mm), bark was thicker (1.75 ± 0.65 mm), pulp yield was higher(57.6%), soluble solids were 9.3 ± 1.3 Brix, and total carotenoids reached 76 ± 4 μg.g-1 b. s. Bee diversity was affected by landscape structure (habitat diversity, cover and isolation from surrounding vegetation), while farm practices produced no detectable effect. Landscape simplification and absence of beneficial farming practices for pollinating bees threatens bee diversity and pollination services delivery. These results highlight the importance of providing bee habitat (floral and nesting resources) within each farm, helping stingless bees by restoring their natural habitats along streams to increase wild bee populations within crops.

Cryptic Species, Sexual Dimorphism, and Taxonomy of Neon Cuckoo Bees genus Thyreus in Thailand

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Taxonomic studies of non-Apis bees in Southeast Asia require intensive revision to reveal the immense diversity of such important pollinators. The “neon cuckoo bee” genus Thyreus (Apidae; Melectini) is a cleptoparasite on larva of common “blue-banded bee” genus Amegilla (Apidae; Anthophorini), an effective pollinator of flowering plants from families Fabaceae, Solanaceae, and Cucurbitaceae. By combining morphological and molecular data (COI gene) from 200+ Thyreus specimens collected throughout Thailand since 1960s, a present taxonomic status, sexual dimorphism, and cryptic species of Thyreus are reassessed. Based on inferred phylogenies using Maximum Likelihood and Bayesian Inference methods, at least eight Thyreus species are recognized, including four new putative species from Thailand. A detailed examination of male genitalia and terminal sclerites correspond with the phylogenetic results. Identification keys to both males and females were constructed for the first time with clear diagnosis characters for both genders. This exemplar work demonstrates the necessity for more future taxonomic research on other native bees in this region for future pollination purposes. A parallel taxonomic study on Thyrues host, Amegilla is currently underway.

Meliponiculture and its potential for pollination in Kerala, India

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Kerala, the Gods own country is having diversified flora and congenial weather conditions suitable for sustainable meliponiculture. It is the art and science of rearing stingless bees for honey production and pollination. Asiatic stingless bees are small sized bees under the family Apidae and sub- familiy Meliponinae. *Tetragonula iridipennis* is the common stingless bees found in Kerala, South India. They lead social life with single queen, few drones and hundreds of workers as in Apis bees. Feral colonies are present in the basements of old buildings, compound walls, tree trunks, hollow blocks etc. The nest architecture includes hive entrance, brood cells, storage pots of honey and pollen, resin dumps, pillars of wax, waste dumps. The All India Co-ordinated Research Project (AICRP) on Honey bees and Pollinators, Vellayani Centre of Kerala Agricultural University has developed technologies for hiving, domestication and management of stingless bees. Stingless bee honey is used in Indian System of Medicine, the Ayurveda for many diseases, even for cancer.

Stingless bees thrive much better in tropical areas and are polylectic. The smaller body size, shorter foraging distance, less aggressiveness, floral constancy, higher longevity of the colonies, efficient worker recruitment behavior towards food sources, medium colony size, lesser swarming tendency, tolerant to high temperature and less pest and disease incidence are the advantages of stingless bees as efficient pollinators mainly in vegetables and fruit crops. Identified 142 plant species, either providing nectar or pollen or both to stingless bees in the State. Out of which, 70 provided nectar alone, 25 provided pollen only and 47 provided both nectar and pollen. They include medicinal plants, plantation crops, condiments and spices, vegetables, fruit crops, field crops, ornamental plants, wild plants and weeds.

Meliponiculture facilitate pollination of various crops which in turn enhance yield and quality seed production. Hence it is recommended to have “one stingless bee colony in each homestead”.

Survey of Insect Pollinators Use for Horticultural Crops in Korea

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We have surveyed the current status of insect pollinators use for horticultural crops in 2016. The use rate and farm number of insect pollinators for 26 horticultural crops were 25.8% and 55,208, respectively. The colony number of insect pollinators used in this survey was 479,777, which include 344,690 for honeybees, 119,104 for *Bumblebee*s, 2,415 for mason bees, 1,317 for flies, and 12,051 for the combination of *Bumblebee*s, honeybees, and mason bees. The use rate of insect pollinators was 59.4% for 11 vegetable crops, such as onion (100%), strawberry (99.9%), oriental melon (93.8%), melon (82.8%), water melon (64.8%), tomato (63.6%), pepper (31.8%), paprika (5.9%), zucchini (1.6%), bitter melon (1.2%), and cucumber (0.2%). The colony number of insect pollinators used for 11 vegetable crops was 449,287, which include honeybees (72.3%), *Bumblebee*s (25.1%), flies (0.2%), and the combination (2.4%) of *Bumblebee*s and honeybees. The use rate of insect pollinators was 9.0% for 15 fruit tree crops, such as mango (100%), raspberry (24.0%), apple (18.3%), persimmon (14.0%), passion fruit (5.5%), boxthorn (4.5%), blueberry (3.8%), Korean raspberry (3.1%), pear (2.7%), cherry (2.2%), pomegranate (1.0%), plum (0.4%), peach (0.2%), kiwi (0.2%), and jujube (0.02%). The colony number of insect pollinators used for 15 fruit tree crops was 30,290, which include honeybees (66.3%), *Bumblebee*s (20.2%), mason bees (8.0%), flies (1.6%), and the combination (3.9%) of *Bumblebee*s, honeybees, and mason bees. Together, farms of 97.8% showed positive effect for the use of insect pollinators and most of farms (97.0%) planed for the continuous use of insect pollinators.

Centridini bees as manageable pollinators of West Indian cherry (Malpighia emarginata DC, Malpighiaceae) orchards

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The West Indian cherry Malpighia emarginata DC (Malpighiaceae) attracted particular interest due to the high ascorbic acid content in its fruits, and some Centridini bees were reported as effectively pollinators of this crop, as the trap-nesting species Centris analis (Fabricius, 1804) and C. tarsata Smith, 1874. The aim of this study was investigated the nesting biology of these two bee species associated with a M. emarginata orchard in Indaiatuba-SP, Brazil, from September 2010 to August 2012. Both species showed a seasonal pattern, but C. tarsata showed a pattern more pronounced in both years ($Z = 40.6$, $p < 0.01$, $r = 0.64$, $p < 0.01$ in the first year; $Z = 24.4$, $p < 0.05$, $r = 0.66$, $p < 0.01$ in the second year) than C. analis ($Z = 67.7$, $p < 0.01$, $r = 0.53$, $p < 0.05$ in the first year; $Z = 89.2$, $p < 0.01$, $r = 0.41$, $p < 0.05$ in the second year). The nesting peak of C. tarsata occurred between October and November in the first year, and in December in the second year; for C. analis, between April and May in the first year and between January and February in the second year. Both species explored 48 plant species of 19 families, being Fabaceae the most representative. The West Indian cherry was the most important source of pollen and floral oil by both C. analis (63%) and C. tarsata (34.1%). The diversity degree of the diet of C. analis ($H' = 1.56$) was significantly lower than that observed for C. tarsata ($H' = 2.38$) ($t = 61.1$, $df = 39491$, $p < 0.001$). While the foraging activity of C. analis showed a low evenness degree due to the high preference for M. emarginata ($d = 0.63$, $J' = 0.47$), C. tarsata explore floral sources more evenly ($d = 0.34$, $J' = 0.67$). The high preference observed in the diet of C. analis and the seasonal pattern observed in the nesting activity of C. tarsata indicate that both species can be used to pollinate M. emarginata orchards.

SYMPOSIUM 15:30-17:30
POLLINATION & FLORA WITH ENVIRONMENTAL CHANGE ROOM 517B

[Lead-off] The role of biodiversity and species interactions in agricultural landscapes and protected areas

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Diverse pollinator communities provide insurance and decrease uncertainty for high quality crop production and ecosystem stability. In diverse bee communities, inter-specific interactions frequently occur and may lead to competition between species changing pollination effectiveness, complementarity in resource sharing and therefore food web structure.
In my talk, I will present results of different research projects ranging from observational and laboratory studies in apple landscapes across Europe and city gardens in Freiburg, a larg-scale manipulation experiemnt of tree diversity to understand bee-parasitoid interactions in forests to honeybee-wild bee competition in semi-field experiments and in a nature reserve in Germany. My overarching research questions are (1) ecological farming and different pollinator ehancement vegetation structures promote bee diversity with variable consequences for crop production; (2) exotic plants in city gardens promote bee diversity in late summer and autumn; (2) tree diversity changes bee-parasitoid interaction webs as higher trophic levels are especially sensitive to tree species loss; (4) inter-specific competition for flower resources occur but honey bee wild bee competition does not necessarily threaten native bee reproduction.
I will end presenting our new bee monitoring project with the United World College and more than 350 participating schools across Germany to answer pressing research questions related to the currently dicussed insect decline.

Agricultural landscapes must include at least 20% of natural habitats: benefits for pollinators and crop pollination

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Governments around the world increasingly acknowledge the need for better policy instruments to enhance natural and semi-natural habitats (NSH) within agricultural landscapes. However, the area of NSH that is required can vary enormously across nations, even when agroecological conditions are similar. This variation reflects complicated political, social, economical, and cultural trends across countries, but also results from the lack of clear guidelines from the global scientific community. Such guidelines are urgently needed, as NSH are quickly decreasing in agricultural landscapes worlwide. Indeed, the destruction of NSH has been identified as a principal cause of lower and less stable crop yields, and the loss of many of nature's contributions that are essential for good quality of life. Here we propose that NSH must comprise at least 20% of the area of agricultural landscapes and discuss the benefits for pollinators and crop pollination.

The campaign 'The Bee, sentinel of the environment': how communities and business stakeholders can help beekeeping

H. Clement
Union Nationale De L'Apiculture Francaise, Paris, FRANCE

As French beekeepers have faced for several years a serious increase of death rate among the colonies, the National Union of French Beekeeping (UNAF) has promoted for the last 15 years an environmental awareness campaign called "Abeille, sentinelle de l'environnement" (1).
This program encourages communities or business stakeholders to protect the bees' environment. Partners sign an environmental charter with several commitments as "Not using pesticide", "Planting melliferous flowers", etc.
This operation turned out to be very successful since more than 80 partners have joined the project: regions, counties, cities or companies. An important number of hives has been set up on buildings or green areas with the support of local beekeepers. The hives establishment and honey harvesting are real opportunities to show people and politicians how important bees and pollinators are.
Every year, UNAF and its partners organize a national event named "APIdays" in more than 120 places in the same time. Open to a large public, this free event helps to present the honeybees' products and to better understand bees' mortalities. In 2018, there were more than 300 press releases on national and international radio, TV, or newspapers.
Beehives in our cities highlighted the beekeeping situation in our countryside and particularly in the area of field crops. With its 15 years of experience, UNAF will be able to provide the beekeepers information and tools to develop such a program in their own countries.
(1)The Bee, sentinel of the environment

The effect of climate variability on bee forage and honey production in Costa Rica

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The flowering period of 53 species of trees was registered for each year between 2012 and 2019 during the dry season (December –

May) in the North West of Costa Rica, an area where more than 200 beekeepers have over 30,000 hives. Two weekly observations were carried out during this period on a specific 173 km long route, visiting the municipalities of Atenas, Orotina, San Mateo, Esparza, Garabito, Abangares, Palma, Mansión, Hojancha and Nicoya. The effect of rainfall and weather phenomena such as hurricanes, and "El Niño" or "La Niña" on flowering period and the average honey production in the area were analyzed. During this period one very strong "El Niño" occurred starting in 2014 and ending in 2016, causing severe drought in the study area, with 44% less than average rainfall. A moderate intensity "La Niña" effect was measured in the year preceding the study period, causing surplus rainfall of over 150 mm (25%) in the study area during the three months preceding the data collection. In November 2016, just after the two-year drought caused by the "El Niño" effect, hurricane Otto caused severe flooding in the study area. It was observed that less than average rainfall in the months preceding the dry season (September-November) caused two to six weeks early blooming of normally late flowering trees, such as Cassia grandis, Byrsonima crassifolia and Bursera simaruba. If the onset of the blooming is compared between years with surplus rainfall (2011, 2017-18) and regular precipitation during these months, no delay in the start of blooming of early flowering tree species, such as Calycophyllum candidissimum, Diphysa americana and Caesalpinia eriostachys, was observed.
In Costa Rica the annual average honey production is about 30 kg per hive. Beekeepers reported exceptionally low honey production in the years 2015 and 2016. On average 30% less honey was reported by 27 beekeepers for 2015 and 55% less during 2016 by 23 beekeepers who participated in the survey.

Apiflora potential of Cucurbita sp. and Cosmos sulphureus for honey production in stingless bees, *Tetragonula pagdeni*

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Microbeekeeping protocol to design a quantitative and qualitative properties of natural honey as taste, aromatic quality, colour, sugar contents as well as some medicinal properties by manage a honeybee flora within the foraging radius of the bees was performed. The aim of this project is feasibility testing of a model in an economically scale. Cucurbita sp. provides nectar by volume 1,250 ± 137 microliter per florescence and sugar content at 43.50 ± 9.30 brix. Cosmos sulphureus provides nectar by volume 15.50 ± 3.40 microliter per florescence and sugar content at 17.25 ± 11.50 brix. Cucurbita sp. and Cosmos sulphureus were two types of flora that were used as signature in honey of stingless bees. Stingless bee honey yield results 3.5 kg/rai which was 45.39 % lower than expected yield. Produced honey was passed qualified the phycio-chemical properties standard of Thailand Food and Drug Administration standard demonstrating that the protocol is highly feasible for beekeepers. The analysis of highly volatile compounds in honey using GC-MS techniques revealed total 34 compounds in honey collected from tested farm consisting of 52.5 % similarity to honey yield from original model.

Pollinator network analysis in agricultural landscape dominated by apple orchards relative to the degree of naturalness in Korea

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Pollination is an very important ecosystem service provided by wild and managed biodiversity in agroecosystems. Recently research in the agricultural ecology has been increasing. Pollination directly affects the yield of crop production as well as the quality factors such as micronutrients or other customer-enchanting properties. We had monitored the pollinators' assemblages on the small-scale crops or wild-flowering plants to see the effect of the land use patterns, measured by the surrounding landscapes. the production of crops and has a significant positive effect on pollinator. In this study, we analyzed the pollination network structure of crop plantation sites in Gyeongsangbuk-do Province and Pyeongchang-gun region in Republic of Korea. The surveys were extensively conducted during was conducted for a month in of August 2015, visiting each plant patches to collect flower-visiting insects with fixed sampling design. The elements of landscape in each region were also investigated quantitatively, and the data on collected pollinator and crops were organized and analyzed together. As a results tTotal of 9021 individuals were collected and registed as species of interactions and 309 species in Hymenoptera, Diptera, Lepidoptera, Hemiptera and so on. were observed. In each region, the crops were mainly produced by Balloon-flower (Platycodon grandiflorum) and perilla(Perilla frutescens var. japonica), while the pollinator was dominated by honeybees(*Apis mellifera*) and some dipterans. Overall, the habitat diversity positively affected the plant-pollinator networks producing more links and depth. Also, there were significant positive association found on the network properties and apple orchard cultivation area. The comparison of the elements of the landscape and the respective collection network level revealed that the pollination of the crops are significantly affected by the increase in the density of orchards around them. The wider the orchard area, the more likely the pollinator abundance and Fisher alpha diversity index showed an increasing tendency. Results imply that the patch-structured perennial crop system could function to harbor pollinator diversity and may possibly be manipulated toward pollinator-friendly habitat such as flower strips to enhance pollination serviceIn conclusion orchard density around of crop plantation had a affect on the pollination network.

POLLINATION AND BEE FLORA

12 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

IDENTIFICATION / CHARACTERIZATION OF FLORAL SOURCES ROOM 517D

[Lead-off] Identification of floral resources used by honeybees and other pollinators

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Melissopalynology, using microscopy to identify pollen within honey, is the traditional method for characterising its botanical composition. An alternative method uses DNA metabarcoding, which involves extracting the DNA found within pollen, sequencing it using next-generation methods and comparing to a reference database in order to identify it. Both approaches yield similar results.
To investigate honeybee plants throughout Europe, we collected data from over 200 papers, covering 25 countries, which used melissopalynology to identify the floral composition of honey. There were clear regional differences, but also key species that were important throughout Europe, with over 50% of papers recording Trifolium and Rubus species.
To examine honeybee plants within one country in more detail, we used DNA metabarcoding to characterise 474 honey samples, collected from throughout the UK. Rubus was the most frequently found genus, followed by the Crataegus, Malus, Cotoneaster group, Trifolium repens, Prunus, Quercus and Acer. We then determined plant use on a monthly basis by conducting a detailed study within the National Botanic Garden of Wales and adjacent organic farmland. Each month, from April to September, all plants in flower throughout the study site were recorded and honey was sampled. The honeybees only visited a small proportion of the available species. In total, 136 plant taxa were found in the honey, with only 16 of these represented by more than 10% of the DNA sequences returned for each month. Foraging was found to change significantly throughout the season, with trees such as Salix, Prunus and Acer being important in April and May. Rubus and Trifolium repens were two major sources in June, July and August, while Hedera helix was a key late-flowering source of forage in September. The greatest proportion of pollen DNA came from native plants, which were supplemented with lower levels of horticultural species.
To investigate plant use by a wider range of pollinators, we used DNA metabarcoding to identify the pollen carried by honeybees, solitary bees, *Bumblebee*s and hoverflies within the Botanic Garden and surveyed hoverflies from plant-species rich pastures in Wales. Again, we saw the importance of Rubus as a key plant for pollinators.

Building Capacity for Integrated Bee Forage Plantations into Multifunctional Land

B. Foster [1], L.N. Lloyd [2], A. McPherson [2], M. Conquer [3], J. Hartnell [1], I. Raine [4], X. Li [4], K. Rogers [4], A. McKenzie [5], I. McIvor [6], T. Jones [6], D. Glenny [7], W. Kaa [8], F. Scheel [4], J.-N. Galliot [2], M. Gonzales [9], T. Roper [10]
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Integrating bee forage plantations to fulfil multiple purposes on different types of land has been the focus of New Zealand Trees for Bees research since 2009. Procedures for designing bee forage plantations that are both balanced and targeted to optimise bee nutrition at the same time as benefiting farming, gardening, honey harvesting and other land uses are based on several streams of research. These include laboratory analyses of crude protein content and fatty acid profiles of pollen, field observations of bee preferences, pollen and nectar profiles of plant species that bees actually use in apiary sites and demonstration of plantations throughout varying geographic regions and climates in New Zealand. Research results are provided to farmers, beekeepers, gardeners and public land managers along with training on how to create bee forage plantations that are low maintenance and relevant to the specific land use operations and goals. Since 2013 our team has installed over 60,000 trees and shrubs on 25 demonstration sites on pastoral, arable, and horticultural farms as well as public land and native bush. The landowners do not set aside land just for bee forage unless the land is solely a beekeeper's home yard for queen raising. Instead, landowners integrate high performance bee plants into their current planting for shade and shelter, erosion control, riparian protection, amenity, pollination support, marginally productive land for honey harvesting and conservation of biodiversity to name a few. The procedures include the use of balanced and targeted flowering calendars, annual bee forage budgets, apiary site pollen profiles and an extensive database with information on over 500 candidate plants with multifunctional purposes. The research has been funded since 2010 by the New Zealand Ministry for Primary Industries Sustainable Farming Fund and apiculture and farming industry participants. Our new grant for 2019 to 2022 provides funding for capacity building via training planting advisers, increasing the diversity, quality and abundance of the best bee plants in the nursery trade and developing online tools to assist with creating bee forage plantation designs for installation in any region and for any purpose or land type in New Zealand (www.treesforbeesnz.org).

The botanical origin of South African honeys: Novel findings and challenges

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Honey is a highly variable natural product that differs significantly between geographic regions with different vegetation types and also between seasons with different floral availability. The demand for honey varieties based on distinct geographic and botanical origin have increased in recent years, with monofloral honeys and honeys with unique properties appealing to elite customers and fetching higher prices on the international honey market. South Africa is a country with very high biodiversity, especially the Cape Floristic Region (CFR) biodiversity hotspot in the Western Cape Province that contains a large variety of unique and endemic plant species. Honeys produced in this unique vegetation type might consequently have their own distinctive characteristics that could potentially increase the value of these honeys. Research into the properties of honey is essential in providing scientifically supported marketing tools to promote exclusive honeys, but is mostly lacking on the African continent. In a novel study on honeys produced within the CFR along the west coast of South Africa, the first pollen library of the area was created and the botanical composition of 67 honey samples was determined through melissopalynological techniques. The honeys produced were mainly from indigenous plant species and honey samples from different apiaries showed some similarities in their botanical compositions - indicating that the honey bees prefer similar floral resources across space. Monofloral honeys were also produced, although inconsistently across honey seasons. Melissopalynology still remains an indispensable method in identifying the botanical composition of honey, however challenges faced when conducting pollen analyses in the CFR, as well as in a broader African context, will also be discussed.

Flowering times of the plants of Khartoum State with reference to their importance for honeybees

S.M.A. Mohamed, E.-I. El-Khidir
University of Khartoum, Khartoum, SUDAN

The survey and evaluation of Khartoum State area for flowering plants lasted for 18 successive monthes. A list of 85 such plants is provided, bee plants abundance and dearth were clearly indicated. The various length of flowering periods of the variuos plant species,floweing density per plant and bees(*Apis mellifera L.* and *Apis florea F.*) preference for some of them were recorded. Indigenous plant species are mostly blooming during autumn and winter seasons.

Exotic plants which are utilized in the area as shade and ornamental plants many of them bloom during summer, therefore, they compensate for lack of flowers in the dearth period.

About dozen plant species can be referred to as good foraging plants (key plants) in the area : Grewia tenax, Ziziphus spina-christy, Balanites aegyptiaca, Acacia nilotica, Acacia seyal, Capparis decidua, Tamarindus indica, Mangifera indica, Albizia lebbek, Plumeria ssp., Eucalyptus ssp., and Acacia mellifera.

The important factor affecting flowering in the area was the availabilty of water (irrigation water, high under ground water-table and rains).

Botanical certification of honey produced in the Caatinga in the Northeastern Brazil

J.E. Melo Nascimento [1], A.J. Souza Pacheco Filho [2], E.G. Girao [3], C. Piffero Câmara [2], G.H. Simoes Pereira [1], V. Arnaut de Toledo [1]
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The Caatinga is an area of approximately 11 million inhabitants, where there are 27 million people living and most of them need of this source of biome for its survival. Throughout its territory, there is a large biodiversity with about 1000 species of plants and 221 species of registered bees. This biome, typically Northeastern, presents special characteristics of floristic diversity, mass flowering in the rainy season and environmental conditions in combination with the presence of the Africanized honeybee, give it great potential for beekeeping activity. The Northeast is one of the largest honey producers in Brazil, although it is considered that beekeeping is still little explored in this region. Honey from the Northeast is appreciated by the most demanding consumers because it has organoleptic characteristics specific to the vegetation. Here, we analyzed the botanical origin of produced honey in Marcelino Vieira town – State of Rio Grande do Norte, Brazil, where a hyperxerophilic Caatinga predominates. For this, samples of honey were collected in two main harvests of January and February; March to April 2016. We verified that the honey originated from the flowering of Croton sonderianus and Croton heliotropiifolius (Euphorbiaceae), known as quince and canopy. Together they account for more than 45% of the total pollen grains found in the first honey harvest, held just over a month after the first rains. As for the botanical certification and the nectariferous evaluation of the samples, it can be stated that honey from the nectar collected from these two native species of the genera Croton during the first annual harvest, represents a monofloral honey. The second annual crop was quite diverse with respect to the botanical origin. Combretum leprosum (27.75%) and Spondias macrocarpa (25.75%) were the main pollen grains found in honey. Another plant that also stood out in the production of nectar was Mesosphaerum suaveolens (2.75%), even though it was represented by few grains of pollen in honey. The honey produced in this period is multifloral, having in its composition the nectar of several apicultural species. The honey obtained represents well the apicultural vegetation typical of the Northeast Caatinga.

What Do Honey Bees Eat? A Quantitative Melissopalynological Study to Determine Honey Bee Foraging Preferences and Influence Local Landscape Decisions

D. Klughers
Bonac Bees, East Hampton, USA

Honey bees (*Apis mellifera*) were introduced to America in 1622 and by the 1950s expanded to almost six-million colonies. Since then, colonies have declined. In the recent past, annual loss has increased to 35% or more. Today, roughly three-million colonies provide $15 billion in annual pollination services to America, and are vital to plant reproduction and biodiversity.
Lack of forage can stress honey bees. It can cause malnutrition and immune dysfunction and promote greater susceptibility to additional health threats. By identifying nectariferous species that honey bees prefer to forage upon, localized landscape practices can be developed to provide honey bees greater food diversity and abundance.
In this study, quantitative melissopalynological analysis was utilized to identify the botanical origin of different honey samples, thus determining preferred honey bee forage. Over a three year period, twenty-eight honey samples were collected from nineteen apiaries on Long Island, New York. Fifteen apiaries were within approximately eighty square miles, and five were 4.5 to 40 miles outside this vicinity. The study area is predominately rural with limited agriculture, constrained by numerous water-bodies.
Analysis established the pollen spectra and percentage in each sample. Sixty-nine distinct pollen grains were identified, comprising 0.40% to 79.5% of pollen spectrum in each sample. Table 1 contains results for the six most common taxa identified.
Table 1.

TAXA	FREQUENCY	RANGE of VALUES	AVERAGE%
Ligustrum (privet)	28/28	1.8% - 79.5%	33.57%
Trifolium/Melilotus (clover)	26/28	2.0% - 30.7%	11.52%
Rhus/Toxicodendron (sumac/poison ivy)	24/28	0.50% - 33.2%	10.67%
Ilex (holly)	23/28	0.90% - 18.8%	7.07%
Cephalanthus (buttonbush)	22/28	0.50% - 19.10%	4.65%
Lagerstroemia (crepe myrtle)	26/28	0.40% - 12.3%	3.62%

These data supports the theory that honey bees prefer to forage upon hedges and trees even when a variety of plants are within their range. Recent research suggests that planting hedgerows and trees, offering efficient foraging with greater food density, helps feed honey bees. Trees and shrubs are an economical and simple alternative to flower plantings. This research project can be utilized by landscape professionals, homeowners, government and others to plant what honey bees prefer. This study can be replicated for other locations.

Pollen analysis and honey physiochemical properties of Gesha-Sayilem Forest in South west Ethiopia

A.I. Merti [1], S. Demissew [2], T. Soromsa [2], K. Wakjira [1]
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Low quality of honey product from different parts of the country leads to high challenges on export market and hence low export earnings. As a part of solution to this problem, investigation on honey samples collected from Gesha and Sayilem districts of the Kaffa Zone, Ethiopia was conducted to identify the quality of the product from the study areas against national and international standards. Assessment of bee forage based on field inventory, pollen load collection using pollen trap and pollen analysis indicated that 79 bee forages with the major pollen count belongs to Schefflera abyssinica, Croton macrostachyus, and Vernonia amygdalina ranging from 66.4 % to 96%, 478.2-58.7%, and, 33.92-64.4% respectively. The results of analyzed honey samples from the areas for moisture, sucrose, glucose and fructose content, shows that 19.5 g/100 g, 69.48 g/100 g, 38.6 g/100 g and 35.5 g/100 g, respectively. Similarly, proline, pH, free acid and HMF values, electrical conductivity and invertase activity of the honey also shows that 210.1 g/100g, 4.05g/100 g, 7 meq/kg, 1.23.87 mg/100g, and 0.16 mS/cm, respectively. Significant correlations were observed between moisture content and electrical conductivity ($r = 0.76$, $p < 0.01$). The study found that all analyzed parameters of the honey samples meet the basic honey quality standards both national and international specifications. From this study, we recommend that potential investors and buyers can create partnership focusing on this large UNSCO delignated Biosphere forest for producing and exporting of quality honey product to the international markets.

SYMPOSIUM 13:00-15:00

CONSERVATION OF FORAGE AND HABITAT FOR BEES — ROOM 517D

[Lead-off] Conservation and design of forage habitat for bees, the challenge of “partial habitats”

N. Williams
University of California, Davis, USA

Lack of reliable forage (pollen and nectar) is a key driver of wild bee declines globally and a major threat to managed honey bees. Wildflower plantings, including hedgerows, flower strips and meadows, are widely implemented to augment existing resources and counteract the loss forage for bees from anthropogenic land use change. I review the value of such plantings as forage, highlighting several empirical studies where we have tested the performance of plant mixes and individual species to support bees and achieve other goals such as benefiting pollination services. I use data on bee visits and traits for individual plants to illustrate a robust framework to help practitioners design plant mixes to benefit bees. I then argue for a more comprehensive view of forage habitat that considers other needs and threats for bees. Bees not only require forage but also sufficient nesting habitat and safety from pathogens and environmental toxins. Despite attention to planting forage, we know little about whether such habitats also bolster nesting or mitigate other threats that may limit bee populations. I used replicated surveys among sites throughout a landscape to demonstrate the benefits of some forage habitats for bee nesting. Including nesting needs can help avoid creating "partial habitats" that may limit the full potential of forage habitat to benefit bees.

Colony health in intensified agricultural landscapes: monitoring the impact of forage availability on honey bee hives in heavily cultivated areas

R.P. Cass [1], A. Dolezal [2], A. Toth [1], M. O'Neal [1], E. Hodgson [1], H. Hendriksma [1], A. St. Clair [1], G. Zhang [1]
[1] *Iowa State University, Ames, USA,* [2] *University of Illinois Urbana-Champaign, Urbana, USA*

Intensive agricultural land-use is frequently cited as a significant contributor to pollinator decline. For example, honey bee colony losses are alarmingly high in regions with high levels of monoculture cropping such as the Midwestern United States. Hive loss in cultivated landscapes is correlated with hive stressors such as poor forage availability and insecticide exposure. However, some flowering crops provide huge blooms of forage to pollinators, and there have been mixed reports about potential costs and benefits of agricultural land to bee health. In this study, we provide a highly replicated, multi-year assessment of honey bee colony growth and nutritional health in Iowa USA, a landscape devoted to the production of corn and soybeans and one of the most intensively farmed areas in the world. The study monitors multiple hive health indicators in colonies placed in conventionally farmed soybean fields. Additionally, the study explores the health benefits of relocating hives from intensified agricultural landscapes to Iowa prairies that have a higher abundance of blooming crops in the late summer period when forage availability is low across the state. The data collected thus far indicate that landscape and time of year have a significant effect on hive health and point to possible strategies for beekeepers with hives in areas of intensified agricultural cultivation.

Foraging distances of commercially-deployed bees: A meta-analysis

R. Miksha
University of Calgary - Department of Biological Sciences, Calgary, CANADA

Three commercially valuable pollinators – honey bees, bumble bees, and leafcutter bees – are routinely relocated by beekeepers to satisfy pollination requirements of some agricultural crops. There is growing concern that movement of commercial bees may impact local native bees through competition, spread of disease and parasites, and by pollinating and promoting invasive plants. To protect biodiversity, managers of conservation reserves often need to know the distance at which commercial bee species engage their landscape. Meanwhile, managers of agricultural crops requiring bee pollination benefit from anticipating the foraging distance of their hired pollinators.
Worldwide, commercially employed western honey bees (*Apis mellifera*), bumble bees (esp., *Bombus terrestris*), and alfalfa leafcutter bees (Megachile rotundata) contribute billions of dollars in pollination value to crops annually. However, we know little about their foraging distances, and the determinants of these distances, which likely include landscape heterogeneity, nest density, colony size, and foraging economics.
I used meta-analysis to obtain flight distances after examining ~400 papers involving these three groups of bees. My primary objective was to determine mean foraging distance for each taxonomic group. Flight distances vary. For example, bumble bees, with ~250 species, have mean foraging ranges that vary interspecifically by an order of magnitude. Overall, mean honey bee foraging distance is over 1000 meters; bumble bees (B. terrestris), ~600 metres; and, alfalfa leafcutter bees, less than 100 metres. I analyze and discuss these results relative to biology and environmental circumstance. These means and their influencers yield important information for both land-conservation and pollinator-bee managers.

The potential of urban agriculture to create landscapes of abundance for native and honey bees

R. Ellis
Western University, London, CANADA

Globally many species of bees are experiencing declining populations and/or health. Cities have been identified by both entomologists and beekeepers as safer places for bees due to increased forage, reduced pesticide use, and a wider diversity of habitats. As a result both native bee advocates and beekeepers in North America are increasingly focused on the urban landscape as a haven for bees.
However, there is a growing division between advocates of native bee habitats in cities and advocates of urban beekeeping. A small body of research has begun to raise questions about whether honeybees pose a threat to native bees by competing with them for sources of nectar and pollen. In response, some native bee advocates argue that cities should use the precautionary principle and discourage urban beekeeping. Urban beekeepers argue that honeybees are an important component of urban agricultural systems and that urban beekeeping may encourage healthier, more resilient honeybees.
Drawing on my empirical research with native bee advocates, urban beekeepers, and urban agriculture practitioners in the cities of Toronto and London (ON) I will argue that urban agriculture initiatives, including urban farms, community gardens, and backyard vegetable

gardens, represent diversified urban landscapes in which both native bees and honey bees can flourish. Using a political ecology analysis of pollination I will consider the questions: How do urban agriculture initiatives nurture landscapes of abundance for bees in the context of growing rural scarcity? Is there potential for native bee advocates, urban honey beekeepers, and urban agriculture practitioners to build alliances to transform local food systems in ways that allow for multi-species flourishing?

Shea tree pollination: patterns and challenges for the conservation of agroforests in Burkina Faso

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The shea tree Vittelaria paradoxa C. F. Gaertner is a renowned multi-purpose tree. Its almonds are the source of shea butter, an essential component of cosmetic and food products and a key source of income for the rural poor. Stable yields of shea require a combination of favorable climatic conditions as well as pollinator density and diversity. In this study, we investigated the diversity of insects associated to shea pollination in two focal regions of Central-South Burkina Faso through the characterization of yields associated to three treatments: open pollination (inflorescences accessible to pollinators), manual pollination (cross-pollination from another tree) and self-pollination (bagged inflorescences). The number of flowers and fruits by inflorescence and by treatment type were determined every 3 weeks until the end of flowering and the fructification. Fruiting and maturation index were calculated and compared, and the results were compared to the patterns of insect abundance and diversity collected by netting, by pan trap collection and by the cup method. The results revealed that fruiting and maturation index from manual pollination was significantly higher than the other pollination treatments. In particular, manual pollination was associated to higher quality and quantity of pollen deposition, leading to a significant increase in the fruiting index and maturation. The study reveals that shea is visited by a large number of insects belonging to several orders. Honeybees (*Apis mellifera*) were the most common pollinators encountered during the field surveys. We will discuss the implications of our findings for crop pollination and biodiversity conservation of shea agroforests. The development of beekeeping will provides lot of honeybees, which will increase pollinating service and then monetary incomes.

Bee-plant interaction knowledge in Brazil integrated through an information system

A.L. Assad [1], K.P. Aleixo [1], S. de Souza [2], D. Canhos [2]
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Pollinators represent a key component of global biodiversity as they play a role in the maintenance of most natural ecosystems and add value to global agriculture. In order to gather and provide information on bee-plant interactions to facilitate decisions on pollination of crops and conservation of bees and plants in Brazil, the Brazilian Bee Studies Association (A.B.E.L.H.A., acronym in Portuguese), in partnership with the Reference Center for Environmental Information (CRIA, acronym in Portuguese), developed the Information System on Brazilian Bee-Plant Interactions. All interaction data were obtained from dissertations, thesis, and scientific articles that surveyed bees on flowers or used pollen analysis as a tool for the identification of plant species used by bees to collect food. Surveys were carried out in many different regions of Brazil and data were collected and organized by the project "Evaluation of the current status of plant-pollinator interactions", coordinated by Prof. Dr. Astrid de M. P. Kleinert of the Institute of Biosciences of the University of São Paulo (IB-USP) with the support of São Paulo Research Foundation (FAPESP). The Information System on Brazilian Bee-Plant Interactions provides data of more than 900 bee species' visits to over two thousand plant species, totaling 17 thousand interaction records. Anyone can search for the scientific name of bees or plants, retrieving a list of plants visited by the bee or a list of bees that visit a specific plant. By clicking on the scientific name displayed in the list, users have access to the reference that documented the interaction. Users can also search for bee-plant interactions at specific phytophysiognomies, environments, or Brazilian states. This data is currently being used by A.B.E.L.H.A. to help develop a more friendly landscape for pollinators and to enhance the quality of areas where beekeepers keep their honey and stingless bee colonies. In addition, as interaction data is based on scientific knowledge and is openly shared online, it can contribute to the development of public policies to protect and conserve bee species, their habitats, and associated flora. The system is accessible to all interested at http://abelhaseplantas.cria.org.br/.

Bee Pasture Design Affects Delay of Disease Spread Among Colonies

J. Rabajante, C. Cervancia
University of The Philippines Los Baños, Los Baños, Laguna, PHILIPPINES

The spread of diseases is one of the major problems in beekeeping. In this paper, we use mathematical models to show that the spatial distribution of forage plants affects how bee diseases are spreading among nearby colonies. In a network of bee colonies, there are cases where transmission of diseases is faster, and there are cases where transmission is delayed. Assuming optimal foraging theory, we show that certain designs of bee pasture are effective in lowering the probability of epidemics. We observe that if two colonies are near to each other, where one colony is infected, the intersection of their foraging area plays a big role in disease transmission. If there are only few flowers in this foraging area, infected and susceptible bees have high chance of encounter. A large patchy foraging area is best for delaying disease spread. This theoretical result is still subject to empirical testing but we suggest that beekeepers already look into the possibility of establishing enough bee pasture, not only as source of food for the bees but also to control epidemics.

BEEKEEPING TECHNOLOGY AND QUALITY

10 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

TECHNICAL INNOVATIONS IN BEEKEEPING ROOM 517C

[Lead-off] Technical Innovations in Beekeeping

H. Evans
Arnia, Newcastle upon Tyne, UNITED KINGDOM

The craft of beekeeping can be considered as both an art and a science; it therefore benefits from technology, as technology applies science to solve problems and extend our abilities.
Historically the evolution of beekeeping technology has been characterised by long periods of steady state, punctuated by significant developments which dramatically change the nature of the craft. The late 1800's heralded the dawn of honey production on a commercial scale thanks to three key technological innovations; the movable frame, smoker and centrifugal honey extractor.
Today beekeeping technology is experiencing another sharp disruption as electronics and information technology, coupled with progress in bee science, help our understanding and abilities as beekeepers. Over the last few years the market has seen a surge in gadgets that offer a myriad of services such as digitised inspection records, remotely monitoring bee hives and assessing pollination efficiency using instrumentation such as hive scales, sensors both in and outside the hive, bee counters and cameras. Beekeepers that benefit from these innovations tend to fall into three distinct groups; while there is an overlap in application, the value extracted from the information is tailored to each.
Hobby beekeepers can track nectar flows, compare colony development year on year and receive automated alerts for broodless colonies, the need to add/remove a honey super, when to feed and now even when the queen goes on her mating flight or if the hive is under attack from Asian hornets! Moreover, the technology offers beginners a helping hand and allows hobby beekeepers to become more intimate with their bees while disturbing them less. Technology has enabled commercial beekeepers to increase efficiency within their operation, improve management practices and increase production. Furthermore, growers can now better track pollination of their crops. Scientists can collect unprecedented volumes of highly granular bee and environmental data simultaneously and precisely coupled with reliable data management.
The question is not if but when will these new technologies become the mainstay of modern precision apiculture, as the rate of adoption will be determined by economic factors and inherently the value these technical innovations bring to all stakeholders.

BeeScanning - monitors bees health

B. Lagerman
Fribi Holding Ab, Lindesberg, SWEDEN

Knowledge when needed to discover parasite varroa, dwv, queen and colony strength. All in your camera.
With a smartphone using the camera and the BeeScanning app, beekeepers can instantly diagnose the parasite mite varroa destructor as well as other features in the hive. Images are taken on living bees on the comb. The tool will also form basis for selection in breeding programmes as well as the basis for population modelling research in the colony. New features are added including brood diseases and bee behaviours, evolving technology using video and sound. Enabling a tool for enhanced knowledge.
BeeScanning is funded by the European Innovation Program, the Swedish Board of Agriculture, The Swedish Agency of Innovations, via Kickstarter and 10 national and international awards.
The app is free to download and free to trial. For now there are 2700 users,10 000 images and more than 10 000 manually annotated regions in 15 classes. View http://tagger.beescanning.com Analysed images are stored and used for furhter training the AI. Images are instantly analysed in the following categories:
Bees with varroa
Queen
Deformed wings
Number of bees
Mean average precision: 83% Recall 67%
Results are presented to the user in absolute figures and as mean varroa infestation level in %. Objects are labeled and displayed to the user for review. Histograms indicates trends of the colony performance as a tool for decisions on treatment and/or breeding purposes.
The Beescanning technology is based on proprietary convolutional neural network, NN, and deep learning. Not by classification or algoritmic image analysing. This means results will continue to improve as the artificial intelligence learns from the ever increasing data its fed by the users.
Metadata as time, geopostion, breed is collected from growing the worlds largest database of images of bees on combs. We aim for a tool that can monitor events, nutritional status, health and make prognoses.

BeeScanning is based on our findings that there is a correlation between the actual varroa infestation level, as measured by alcohol washing or Apistan, and varroa that is optically detectable. https://beescanning.com
Björn Lagerman, founder Fribi Holding AB

High Tech Research leads to Grass Roots Methods to prevent Global Honeybee Losses

M. Greco [1], B. Coates [2], E. Feil [2]
[1] *Charles Sturt University, Wagga Wagga, AUSTRALIA,* [2] *University of Bath, Bath, UNITED KINGDOM*

The widespread decline of honeybees globally has serious consequences for ecosystems and agriculture. Bees are the major insect pollinators and thus mitigating their declines is of major importance to global food security. Recent findings, during a high resolution diagnostic radioentomology study, indicated that honeybees show preferences when storing food and when feeding other bees. If this is indeed the case, then honeybees might also preferentially spread pathogen, which is in the food, to other bees within their hive. Here we show that bees from certain hives show preferences while feeding other bees and that bees from other hives do not. The simple, new method developed for assessing food and pathogen transmission in bees will help beekeepers to select and breed bees that have a lower propensity for spreading disease within a hive. Therefore, the beekeepers' own selection and breeding programs will help mitigate bee declines, at the grass roots level, globally.

Pollination Hive Grading Using Infrared Image Analysis

E. Symes, W. Wells, G. Riggs, G. Alexeev
The Bee Corp., Bloomington, USA

The Bee Corp (TBC) received a Small Business Innovation Research (SBIR) Grant from the National Science Foundation (NSF) to research and develop a beehive grading solution for commercial pollination. TBC's Phase I research proved the viability of infrared (IR) image analysis to determine hive population from a commercial and a technical feasibility perspective. Modeling hive population from IR images included image processing and segmenting, statistical modeling, feature engineering, model optimization, and performance testing. To build these models, ambient, biological, and thermodynamic factors were included to account for heat effects on the IR image not caused by the bees. TBC then built an image recognition model to find the hive in each image in order to automatically extract thermal values for analysis. During Phase I, TBC proved the technology is viable and IR data can be used to accurately calculate colony size. TBC also demonstrated market demand and commercial viability for the solution. Through these pilot tests, TBC determined necessary improvements for its NSF SBIR Phase II grant application for full commercial scalability and viability.

IoT applications for honey bee colony condition - what's the buzz all about?

F. Edwards Murphy
ApisProtect, Cork, IRELAND

Contributing $174 billion worth of pollination to the agri-food industry annually, honey bees play an essential role in global food production. However, many countries are experiencing extensive honey bee loses, in the U.S., commercial beekeepers reported declines of 38 percent of colonies in 2015-2016.
ApisProtect is an Irish agtech company established to help reduce honey bee losses worldwide. ApisProtect uses a unique combination of sensors to monitor honey bees in the hive and collects temperature, humidity, CO2, sound, and movement data from a single sensor unit. The key value of this technology is the processed data – a high level overview of each apiary with a breakdown of which hives are doing well, which ones are likely to experience problems, and which hives currently need immediate attention providing a 24/7 early warning system to help reduce honey bee losses. These sensors will help beekeepers identify a wide variety of problems, earlier than they can using traditional inspections.
Results from global trials of the ApisMonitor units will be presented.

BeeKing a digital assistant for efficient beekeeping

J. Kronbergs
Latvian university of agriculture, Jelgava, LATVIA

It is not easy for beekeepers to take records and collect data digitally during inspection of beehives with the traditional way of data entry. Beekeepers also have a fragmented knowledge base, making learning from other beekeepers experiences difficult or time-consuming. By introducing Speech to Text (S2T) interface, beekeepers are able to take digital apiary records and plan their activities, check the results, and ask for support or share their knowledge within the same service platform called "BeeKing".
The idea about the project came to me as a beekeeper with professional background in business process re-engineering in August 2016. A strong presence of machine learning and neural computing competence center for S2T and Text 2 Speech (T2S) technologies in Latvia made possible a dream of having voice based hive records. In January of 2017 together with Latvian Beekeepers Association we did a

survey to understand beekeepers' needs and problems concerning apiary records and their use during inspections. We also collected data about openness of beekeepers to new, smartphone-based technologies for use during inspections and ways how beekeepers are learning. We got quite a high response rate with more than 20% of participants willing to apply for early testing of such a tool. After gathering feedback from users and gaining clear understanding of their needs, we developed the project idea and in 2018 minimum viable product was ready to use and made available to beekeepers being interested in "trial" use of it.
More beekeepers than expected applied for a trial period use.
From 2019 a training program was established together with Latvian Beekeepers Association and all beekeepers can join it to learn more about hive records practices, as well as to try voice-based hive records with BeeKing. As result of project Latvian Beekeepers become more interactive within community and responsive to Latvian Beekeepers association.
BeeKing service platform is multilingual, currently in Latvian and English. It currently is being developed towards deeper use of artificial intelligence to enable efficient beekeeping.
For more information about our application see here: https://beeking.eu/en/index.html and in recent days it will be published also here: https://enrd.ec.europa.eu/projects-practice_en

Water balance control in the bee colony

V. Semeniuk, N. Semeniuk
Union of Beekepers of Ukraine, Kyiv, UKRAINE

Excessive metabolic moisture in the hive during the wintering period is the main enemy of the bee colony. Moisture accumulates in the hive as a waste product of bees, which consume honey and, thus, receive energy to maintain thermal comfort in the colony.
Offered, without ventilation remove excess moisture from a bee nest, when metabolic moisture moves freely through the capillaries of thermally insulated material. The direction of movement of moisture always occurs from a more humid to a drier zone and does not depend on the direction of the gravity force.
A device has been developed that applies this mechanism practically, namely: (i) condenses moisture in the material; (ii) moves moisture beyond the limits of the bee's nest; (iii) collects moisture in the zone waterproofed from the bee nest; (iv) gives the bees accumulated moisture to prepare food in early spring after the flight, when departure conditions are not always favorable.
When preparing for wintering, the device placed in the hive instead of the two last frames, since it has the same dimensions as a bee frame.
Device was tested in beehives of the apiary during the wintering under the open sky in conditions of the middle geographic band (Kyiv region, Ukraine), when the wintering period was five months without flying. After wintering, almost all dead bees removed from the hive already at the first cleansing flight. This shows the device effectiveness to create the comfortable wintering conditions for bees.
Application for Patent of Ukraine on this device is pending.

SYMPOSIUM — 13:00-15:00
CHARACTERIZATION OF BEE PRODUCTS — ROOM 517C

[Lead-off] Chemical characterization of propolis: good news, bad news

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Propolis (bee glue) is the most fascinating bee product, important element of the social immunity of honeybees and a valuable medicinal product, used for centuries as a remedy in the traditional medicine of numerous nations. Modern science has proved its diverse pharmacological activities: antimicrobial, antioxidant, antiinflammatory, immunostimulating, hepatoprotective, cytotoxic, etc. The chemical composition of propolis and its biological properties are determined by the plant resins which bees collect for its production. As bees inhabit almost all ecosystems all over the world, propolis chemistry depends on the local flora. The result is the existence of numerous chemical types of propolis, the most popular being poplar (European) propolis, and green Brazilian propolis. However, researchers continue to discover new propolis chemical types, containing new potent bioactive constituents. In recent years, stingless bee propolis is gaining popularity, and it turns out that its chemical diversity is significantly larger than that of *A. mellifera* propolis. This diversity makes propolis chemical studies challenging but also rewarding, as they bring new knowledge of the abilities of bees to survive, of bee-plant relationships, and of new bioactive molecules. During the last few years, we have studied propolis from *A. mellifera* and different stingless bee species, and from different geographic regions (North and South America, Pacific islands, South-East Asia) and identified a number of potent antimicrobial and antioxidant compounds: diterpenes, triterpenes, stilbenes, xanthones, homoisoflavanes, among them new natural molecules and such new for propolis. These findings strongly support the idea that propolis should be chemically characterized before any biological tests, and propolis could be standardized only if its chemical type is known. We suggest a way to rapidly recognize known propolis types (dereplication) by GC-MS analysis of propolis extracts using a home-made database of chemical profiles and chemical markers of different propolis types. Our results demonstrate that good level of chemical reproducibility for industrial purposes could be achieved through geographical selection.

Metabolomic Based on Gas-chromatography-mass Spectrometry Analysis Reveals the Differences in Volatile Metabolites of Royal Jelly from different honeybee stocks

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Royal jelly (RJ), a glandular secretion mainly produced from the nurse bees, is vital for determination of a female larva's fate developing into reproductive queens and sterile workers and for human health promotion. The differences of volatile metabolites of RJ samples produced by different honeybee stocks, however, are poorly understood. The volatile metabolites of three RJ samples from high and low honeybee stocks with different genetics in RJ production were extracted by headspace-solid phase micro-extraction (HS-SPME) and analyzed by gas-chromatography-mass spectrometry (GC-MS). The results of principal component analysis (PCA), hierarchical clustering analysis (HCA) of the volatile metabolites in three RJ samples are suggestive of the fact that honeybee stock selected increasing RJ (RJBs) for almost 40 years has shaped distinct volatile metabolite profile compared with the unselected counterparts, Italian bees (ITBs). Among the 38 differential metabolites identified, most were quantitatively different rather than qualitatively. Our hitherto in-depth volatile metabolite is the first report that characterizes the volatile metabolites in RJ samples and is helpful to gain a novel understanding biochemical nature of RJ.

Chemical composition of Castanea sativa and polyfloral bee pollen from Slovenia

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Bee pollen contains different nutrients which are important in human nutrition and several studies confirm that bee pollen has antioxidant, anti-inflammatory and antimicrobial qualities. Nutritional composition of bee pollen varies depending on its botanical and geographical origin, season of collecting, environmental conditions and beekeeping practice. The aim of this study was to characterize fresh bee pollen from Castanea sativa (n=10) and polyfloral bee pollen (n=36). Bees mainly collect polyfloral pollen, which is a mixture of different botanical origins. Slovenia is a region where chestnut trees (Castanea sativa) are one of the main sources of nectar and pollen for bees. If the weather conditions are good, beekeepers can collect chestnut bee pollen during blooming season. In bee pollen samples, contents of water, protein, fat, dietary fibre, ash, amino acids and minerals were determined and the total carbohydrate content and energy value were calculated. Before the analysis, the botanical identification of bee pollen was carried out. Our data were compared to the standard of the international basic composition requirements for bee pollen in human nutrition. Significant differences were discovered in protein, fat, and individual amino acid content and in minerals between these two bee pollen origins. With multivariate statistics, we determined the parameters that are significant for analyzed bee pollen and could serve to classify samples into groups. The use of bee pollen in human nutrition and even in therapy demands standardization of its chemical composition and establishment of quality criteria. The presented data serve as an indication of nutritional quality of Castanea sativa and polyfloral bee pollen from Slovenia.

Antioxidative and Anti-inflammatory Activities of Ethanol Extract of Geopropolisfrom Stingless Bee *(Heterotrigona itama)*

S. Liping, W. Bei, W. Kai
Institute of Apicultural Research, CAAS, Beijing, CHINA

Stingless bees (Meliponinae) are one of the important pollinators in tropic and subtropical area, differs from *Apis mellifera ligustica* with typical stingless characteristic.Stingless bee collected more propolis than *Apis mellifera ligustica*.Nevertheless, here is lack of pharmacological investigations on geopropolis collected from stingless bees. This study aims to evaluate the in vitro antioxidant and anti-inflammatory activities of the ethanol extract of geopropolis collected by stingless bee – *Heterotrigona itama*, which is an indigenous stingless bee specien Malaysia. Total polyphenolic and total flavonoids compounds in the EEHI was measured by Folin-phenol method and AlNO3 colorimetry. The antioxidant activity was investigated using DPPH and ABTS free radical scavenging assays. This study using lipopolysaccharide (LPS)-stimulated murine macrophage (RAW 264.7) inflammatory model, to evaluate the in vitro anti-inflammatory effects of EEHI. Based on no cytotoxic concentrations, we measured effects of EEHI on NO release, and on the expression of inflammatory factors (IL-1beta,IL-10 and INOS) as well as antioxidant gene (HO-1) expressions in LPS-activated macrophages. We further taking nuclear factor-kappa B signaling pathway as a breakthrough point to investigate the potential anti-inflammatory mechanism of EEHI. Results showed that contents of total phenolics and total flavonoids in the EEHI were 54.70 ± 0.65 mg(GAE)/g and 116.20 ± 0.12 mg(QE)/g, and the IC50 values of the EEHI for scavenging of DPPH and ABTS free radicals were 275.60±0.22 ug/mL and 284.00±0.01 ug/mL, respectively. In LPS-stimulated macrophage cells.EEHI significantly inhibited the release of NO and the expression of pro-inflammatory cytokines genes (IL-1beta, IL-10 and INOS), and increased the expression of HO-1 gene in a dose-dependent manner. Further study

showed that EEHI significantly inhibited the phosphorylation of NFKBIA protein and nuclear migration of nuclear factor-kappa B-p65, suggesting that EEHI have potential anti-inflammatory effects by inhibiting the activation of nuclear factor-kappa B signaling. In conclusion, the ethanol extract of geopropolis collected by H. itama is abundant in polyphenol, has good antioxidant and anti-inflammatory effects, which has great potential for exploitation and utilization.

Comparison of physicochemical and bioactive properties of turkish pine honey and New Zealand manuka honey

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Altiparmak Gida San. Ve Tic. A. S. Research Center, Istanbul, TURKEY

Pine honey is a valuable honeydew honey which is produced in Turkey with 90% majority. Elucidating the dominant properties of Turkish pine honey and New Zealand manuka honey is the main objective of this study. The level of 43 different flavonoids and 13 minerals were determined as well as moisture, acidity, conductivity, diastase, invertase, proline, colour, HMF, sugar profile, total disaccharides and antimicrobial activity of the honey samples.
Methylsyringate, shikimic and quinic acid were the most abundant flavonoids detected in both varieties. Phenyllactic acid and t-cinnamic acid were detected only in manuka honey. Amount of protocatechuic acid in Turkish pine honey was significantly higher than manuka honey. Moisture and acidity values of both honey types were similar while the value of the diastase and invertase of pine honey were approximately two times higher than manuka honey. The average conductivity value of Turkish pine honey is 1.12, while this value was 0.5 in Manuka honey. Total disaccharides of manuka and pine honeys were 5,7% and 9,4% respectively. It was determined that the average amount of potassium in pine honey and manuka honey were 2866 mg/kg and 1095 mg/kg respectively. The antibacterial activities of Turkish pine honey and Manuka honey were investigated against the reference strains using microdilution method. The Minimum Inhibition Concentration (MIC) of Turkish pine honey against S.aureus varied between 3,12% and 25%, while the Manuka honey changed between 6.25% and 25%. The MIC values of the honey types against E.coli ranged between 12.5% and 25% (w/V). According to the study Turkish pine honey showed higher antibacterial activity than manuka honey (UMF 10 +) against S.aureus and E.coli reference strains.
According to comparable analysis parameters, Turkish pine honey has better values of potassium, iron, zinc and protocatechuic acid while it has similar values of antioxidant capacity, methylsyringate and shikimic acid concentrations versus Manuka. Manuka honeys with UMF values above 15+ showed higher antibacterial activity than Turkish pine honey against S.aureus and E.coli reference strains.
The results showed that Turkish pine honey is equally valuable as Manuka honey.

Geographical characterization of monofloral citrus honeys by nuclear magnetic resonance and chemometrics

M.G. Tamaño [1,6], A. Lopez [2], A. Bonini [3], G. Danners [4], G. Fagundez [5], A. Godoy [3], G. Moyna [2], L. Fariña [3], E. Dellacassa [3]
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Being honey one of the most studied foods by its nutritional and medicinal properties, it also is one of the matrices whose components represent a chemical challenge due to its diverse origins (floral, chemical modifications in the hive). Usually the floral origin of honey is determined by pollen analysis (melyssopalinology), since it can reflect the source of nectar. However, this approach has the limitation of needing specialists. In addition, some pollen grains are underrepresented in honey pollen, such as those of Citrus species, where a minimum of 10% of Citrus sp. in pollen is enough in order to consider a citrus honey as monofloral.
In this study we present the investigation of a combined NMR and chemometric data analysis approach to describe the variability in the composition of citrus honey samples and to discriminate samples from different geographic (Argentina and Uruguay) and botanical origins (native flora).
Forty-honey samples obtained from flowers of citrus and different other plants were studied. All samples were provided by the beekeepers and stored at room temperature (18-23 °C) from the time of acquisition to spectral analysis (max. six months).
Honey samples were dissolved in CDCl3 with 0.03% TMS as internal standard. The 1H NMR experiments were recorded at room temperature using a a Bruker AVANCE III 500. Phase correction was performed manually for each spectrum and the baseline correction was applied over the entire spectral ranges. A database consisting of spectra from authentic samples describing the regular range of product variation was built, the spectra were normalized to the total area excluding the solvent zone and the data were exported to perform the multivariate statistical analyzes (PCA, PLS). These methods were compared with objective to determinate the classification model that shows better prediction ability.
The application of chemometric methods to 1H NMR spectra allowed to discriminate the citrus honeys produced in the Provinces of Concordia (Argentina) and Salto (Uruguay). Moreover, the chemometric methods for pattern recognition showed that it is possible to classify the commercial honey samples according to their volatile aroma profile identifying the type of compounds involved.

Characterization of Indian bee pollen

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The present investigation for the first time aimed to characterize the Indian bee pollen from South-western, North-western and Northern regions of India collected during Jan-Sep 2017. The palynological analysis identified the Brassicaceae, Poaceae and Asphodelaceae as predominant botanical families of bee pollen and nutritional composition reported the amount of carbohydrate, crude fat, protein, ash content, and crude fiber as 35.42–46.13%, 4.85–9.39%, 16.65–25.14%, 1.97–3.04% 3.24–5.72%, respectively. The high amounts of omega 3 fatty acids were recorded with first-ever detection of eicosatrienoic acid, Gamma-linolenic acid, eicosapentaenoic acid, heptadecenoic acid, and dihomo-gamma-linolenic acid. The amino acid composition revealed the presence of all essential amino acids whereas the mineral analysis showed the exceptionally high amount of iron (243 mg/kg) in pollen. The pollen samples from all botanical origins contained the highest concentration of niacin (18.42-31.35 mg/kg), panthothenic acid (3.76-11.42 mg/kg), and vitamin E (1.63- 9 mg/kg). The high amount of proteins, essential fatty acids, amino acids, and minerals suggests the potential of Indian bee pollen as a functional food. Principal component analysis identified the principal components to differentiate the bee pollen based on botanical origin and cluster analysis categorized the pollen samples into three groups successfully.
Keywords: Bee pollen, botanical origin,omega-3- fatty acids, essential amino acids, mineral composition

SYMPOSIUM — 15:30-17:30
DETECTION OF RESIDUES IN HIVE PRODUCTS — ROOM 517C

[Lead-off] Contamination of world honey by neonicotinoids - what implications for beekeepers?

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The worldwide decline of pollinators is threatening the ecosystem services they provide (pollination) and the biodiversity that depends on them. Although the responsibility of this decline is nowadays largely believed to be attributed to neonicotinoid pesticides there was no study at the global scale that allowed to measure the exposure of bees to these substances.
In a study published in October 2017 in the journal Science, the University of Neuchâtel and the Botanical Garden of Neuchâtel assessed the global exposure of pollinators to neonicotinoids by analyzing 198 honey samples from across the world.
We found that 75% of samples contained at least one of the five tested compounds (acetamiprid, clothianidin, imidacloprid, thiacloprid and thiamethoxam). Forty five percent of samples contained two or more compounds, and 10 % contained four or five. This is particularly alarming as the coexistence of neonicotinoids and other pesticides may increase harm to pollinators.
Our results confirm that the majority of bees are exposed to neonicotinoids throughout the world. Although measured concentrations are below the levels believed to be harmful for human consumption (average value: 1.8 ng/g), nearly half (48%) of the samples exceeded the value of 0.1 ng/g for which a negative effect on bees has been demonstrated.
This study therefore does not put into question the quality of honey, but it is alarming for bees - and therefore for beekeeping - because it shows that most pollinators are exposed to pesticides. Based on this study it appears that the use of neonicotinoids is not compatible with the long-term maintenance of bee populations: this encouraged some beekeepers to demand the ban of this category of pesticides.
From a socio-anthropological point of view, we analysed the trajectory of our paper and of our pollical engagement in scientist, beekeeping and politician arenas. We showed how difficult it can be to be politically engaged as a scientist or as a beekeeper on a particularly political question.

Why does New Zealand's high activity Manuka honey fail AOAC C4 sugar tests

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New Zealand's high value manuka honey is one of the most famous honeys in the world due to its unique non-peroxide anti-microbial properties. However this honey is prone to fail one of the major tests for adulteration; the AOAC 998.12 C4 sugar test for cane sugar or corn syrup additives.
Over the last 10 years, millions of dollars' worth of honey exports have been seized, destroyed or returned to its origin, by border testing officials because of non-compliant C4 sugar test results.The AOAC 998.12 method prescribes that honey is considered pure when the test results is less than 7 % of C4 sugar. However for Manuka honey, a large number of false positive failures occur (when the honey is known to be free from sugar adulteration, yet it still fails the test). The rate of failures is especially prominent for high quality Manuka honey, such as those with methylglyoxal (MGO) content (above 200 mg/kg).

A number of studies have been undertaken to investigate possible causes for this pre-disposition of Manuka honey to have false positive fails. Given that it is a highly sought after product, the barriers preventing the highest quality Manuka honey passing this test need to be addressed and the test amended to account for the naturally occurring processes which affect Manuka honey

We propose a mechanism of the change in carbon isotopic composition of Manuka honey's protein, which might explain the high rate of false positives in the C4 sugar test.

Search for exposure gradients to certain atmospheric pollutants by biomonitoring foraging honeybee as an indicator of socio-environmental inequalities between different neighborhoods of Québec City

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Université Laval, Québec, CANADA

Social inequalities in health are evident between different areas in Québec City, as shown by the 6-year difference in life expectancy from one part of the city to the other (Direction de santé publique du CIUSSS de la Capitale-Nationale, 2018). This public health problem is influenced by social factors as well as environmental ones. Among them, the air contamination has an impact on vulnerable populations who are especially sensitive to air pollution due to a synergetic and additive impact of multiple risk factors like psychosocial and environmental stressors resulting in a lower capacity to cope with environmental hazards exposures (Pratt et al. 2015, Morello-Frosch et Shenassa, 2006). The size and the number of air masses along with the low number of sampling stations only make possible a superficial exposure discrimination in the city. This situation justifies the need for a new research method. So, we suggest the use of an innovative zoonotic biosurveillance: the use of honeybees as environmental biomarkers, which has been largely documented mostly in Europe (Perugini et al. 2009, Lambert et al. 2012, Gutiérrez et al. 2015). Bee sampling will make it possible to compare the air pollution from one neighborhood to another and to establish the exposition gradient between them. To verify this thesis, hives will be placed at seven sites determined by their environmental and social characteristics, as well as their proximity to physicochemical stations. Every three weeks, about a hundred bees will be harvested from each site, then will be analyzed to check the presence of heavy metal (As, Ni, Pb and Cd) and Polycyclic Aromatic Hydrocarbons (PAH). These results will be spatiotemporally compared to those from the physicochemical stations, to verify if the two monitoring methods show similar tendencies. Finally, biomonitoring results will also be compared to evaluate the bee potential to reveal an exposure gradient to contaminants and so constitute new social environmental inequalities indicator between different sectors of Québec City.

Pesticide residues in beehive matrices are dependent on collection time and matrix type but independent on proportion of foraged oilseed rape nor agricultural land in foraging territory

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Bees as valuable pollinators of crops and wild plants encounter numerous toxicants throughout their lifetime. In agricultural landscapes pesticides are commonly used and while bees prefer abundant food sources the probability for pesticide residues to end up in beehives is high. Using honey or bee products as indicators for environmental pollution has been proposed now and then. Often people also think that the proportion of agricultural land in bee foraging territory determines the amount of pesticide residues bees encounter. However, defining bee foraging territory is not easy because this is constantly changing depending on flowering conveyer, other bee competitors and weather conditions.

The aim of our study was to clarify whether different beehive matrices reflect similar pesticides residues, which bees gather with food from their environment, and how are these correlated to land use types in foraging territory and forage preferences.

We tested bee collected pollen, beebread, honey, nurse bees and honey bee larvae for presence of concurrently used agricultural pesticides in Estonia. The samples were collected at the end of May and mid-July to include oilseed rape – the main honey crop in northern climate. We found that different sample types contained very variable composition of pesticide residues, which were different also by collection month and year. The result, which is easily explicable by the changing need for agricultural pesticides. However, pesticide residues did not correlate with the proportions of cultivated land in foraging range. The composition of pesticide residues in different samples were variable and were not correlated with oilseed rape pollen in forage.

Elevated coumaphos residues in beeswax resulting from application of CheckMite®

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Since more than 25 years, we monitor residues of acaricides in commercial Swiss beeswax. In the recent years, we noticed an increase in coumaphos residues. Hence, we were wondering if this could be due to the use of CheckMite®, a product that is authorized in Switzerland for beekeeping since 2007. To test our hypothesis, we treated 15 *Apis mellifera* colonies in the autumn with CheckMite® according to the instructions of the manufacturer and subsequently studied the distribution of coumaphos in beeswax. Residue levels were highest immediately after treatment, especially in wax of brood combs that was in contact with the CheckMite® strips. During the following spring season, the coumaphos concentrations ranged between 36 and 159 mg/kg in wax of brood combs with contact to the CheckMite® strips. Residues were about 10 times lower in wax from combs without contact. Coumaphos was also detectable in wax that was not present during the treatment, such as newly constructed wax, wax of honeycombs and capping wax, respectively. In conclusion, the treatment of honeybee colonies with CheckMite® containing 2.72 g of coumaphos per application led to substantial residue levels, especially in the central combs with contact to the strips. Therefore, beeswax should not be recycled after CheckMite® application to prevent elevated residue levels in new foundations. On the other hand, we consider it as highly problematic to discard beeswax, since there is already an existing shortage of recycled beeswax for the production of new foundations.

Sugaring-out assisted liquid-liquid extraction for the determination of contaminants in bee products

W. Chen, S. Wu, Z. Gao, X. Tu
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Sugaring-out assisted liquid-liquid extraction (SULLE) is a novel sample preparation method which shows advantages of simple, rapid, low-cost, and environment-friendly. In this presentation, we will systematically discuss the parameters of SULLE [1], and exhibit the applications of SULLE for the determination of bisphenol contaminants in royal jelly [2] and the analysis of neonicotinoids pesticides in honey sample.

Pesticide confrontation - Are nurse bees able to protect their brood

K. Wallner, F. Boehme
University Hohenheim, Apiculture Institute, Stuttgart, GERMANY

As pollen is an important ingredient to produce larval food, a contamination with pesticides could entail severe consequences on the colonies well-being. However, the fate of pesticides originating from the pollen during this process is unknown. We designed two experiments to trace possible pesticide residues in royal jelly (RJ) as well as in worker jelly (WJ) back to the protein source. We conducted two field experiments with free flying honeybee colonies where we fed a mixture of commonly found pesticides mixed in high concentrations (34.0-9021.8 ?g/kg) into a pollen-honey diet. While feeding, we initiated a queen rearing within the colony to obtain RJ, presumably contaminated with the given pesticides, in the first experiment. In the second experiment, worker larvae were reared during the time the contaminated pollen diet was offered. WJ was harvested on four successive days from larval age three to six. RJ and WJ were subjected to a multi-residue analysis. Seven (out of 13) substances were rediscovered in traces in the RJ. In WJ samples, 6-12 substances (out of 13) were detected in increasing concentrations depending on larval age and pesticide. The increasing number of pollen grains in WJ of older larvae seems to be responsible for the increasing amount of pesticides detected in the WJ samples.

SYMPOSIUM 10:00-12:00
QUALITY CONTROL & FOOD SAFETY ROOM 517C

Standards And Analysis Methods For Honey Quality Control And Food Safety

W. Reybroeck
ILVO (Flanders research institute for agriculture, fisheries and Food), Melle, BELGIUM

Honey is a natural and healthy food product that needs to fulfil quality and food safety standards. Some standards are related to the intrinsic quality, composition and authenticity of honey, while other legislation is setting standards for residues of veterinary drugs and for contaminants like pesticides and heavy metals. Honey must also meet microbiological quality criteria like for osmophilic yeasts, xerophilic moulds and spores of Clostridium botulinum and respect GMO (genetically modified organisms) legislation. Other health issues like pyrrolizidine alkaloids in honey are not regulated so far. Finally other specifications like for the pollen spectrum or organoleptic features could be applicable.

On global scale there exist a FAO Codex standard for honey which Annex is intended for voluntary application (international recommendations) by commercial partners and not for application by Governments. In the European Community, the Council Directive 2001/110/EC of 20 December 2001 relating to honey is laying down criteria for composition, quality and labelling of honey.

Member States shall, whenever possible, use internationally recognised validated methods of analysis to ensure compliance with the compositional characteristics and additional specific statements for all honey marketed in the Community. The International Honey Commission (IHC) was publishing a compilation of harmonised methods. However this edition is limited to intrinsic honey quality and honey composition parameters.

Performance of analytical methods for propolis: an interlaboratorial trial of the international honey commission

M. Lopes [1], L.F. Nunes [1], S.I. Falcão [1], A. Pereyra [2], C. Kunert [3], G. Beckh [4], H. Schreiter [5], O.G. Çelemli [6], K. Sorkun [6], S. Georgé [7], L. Paulo [8], S. Gardini [9], M.T. Sancho [10], V. Bankova [11], T. Dastan [12], T. Tananaki [13], M. Vilas-Boas [1]

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The use of propolis as a raw material for cosmetic, pharmaceutical or even food applications requires a standard definition of its composition, no matter what differences may be found between samples produced on different parts of the globe. This is a subject of long debate in the scientific community and now it became a new topic for discussion within the International Organization for Standardization (ISO), in the recent established subcommittee ISO/TC 34/SC 19 “Bee Products”.

The accomplishment of these goals requires previously the definition of standard methodologies that could be use to describe the product properties but at the same time can be widely applied with precision and feasibility.

With this problem in mind, the Propolis Working group of the International Honey Commission designed a collaborative study to harmonize basic analytical methods and evaluate their accuracy and robustness. Doing so, the group looked to strength the scientific knowledge and establish a background for future definition of propolis standards for industry, producers and laboratories.

The collaborative study was accomplished by 12 laboratories from 9 countries and a company experienced in propolis trade. The samples under analysis, representing fifteen different origins around the world, where fractionated and distributed to each laboratory, and so all participants analysed the same material. Overall, six parameters were evaluated: ash, wax and balsamic extract on raw propolis, and total-phenolics, flavones and flavanones on propolis extract.

The results were evaluated following the international guidelines ISO 5725 [1]. Outliers were removed using Cochran’s test, to check the homogeneity of variances at certain levels, and Grubb test’s, to check the consistency of the laboratories average. The outputs revealed good reproducibility for ash, wax, balsamic content and flavones, while for flavanones the method is clearly unsuitable. Additionally, it was possible to confirm that the use of ultrasounds as alternative method for extraction requires further developments.

Acknowledgment: This work was financed by the Rural Development Program 2014-2020, PDR 2020, through the project DivInA, PDR2020-101-031734.

References:

[1] ISO 5725, 1994. Accuracy (Trueness and Precision) of Measurement Methods and Results.

A survey on the authenticity of *Apis cerana cerana* honey in Chinese retail market

J. Tian, Y. Zhang, J. Si, C. Zhang, H. Zheng, F. Hu
College of Animal Science, Zhejiang University, Hangzhou, CHINA

Honey is a natural healthy food that is loved by people all over the world. In Chinese retail market, the price of *Apis cerana* cerana honey (ACH) is usually several times higher than that of *Apis mellifera ligustica* honey (AMH) due to the production limit of the former, leading to wide adulteration and counterfeiting of ACH by AMH. Through constant research and study, our team has established a system for identifying the two honeys. As little as one percent incorporation of AMH in the mixture can be detected by this indification system. Indicators used for identification were proteins, hydrocarbons and gene components. In this survey, we used the identification system to test the authenticity of ACH in Chinese retail market. A total of 81 commercial ACH samples were tested. The results showed that among all the 81 samples, 67 were AMH, 12 were ACH and 2 were pure syrup. The proportion of real ACH was only 15%. Samples of ACH mixed with AMH were not detected. The results showed that ACH was seriously counterfeited in Chinese honey market.

Major Royal Jelly Protein 4: A possible Fresh Marker for Royal Jelly

H. Hu, J. Li, C. Ma, M. Feng, Q. Wei
Institute of Apicultural Research, Chinese Academy of Agricultural Sciences, Beijing, CHINA

Royal jelly (RJ), a yellowish white, creamy liquid, is the food of queen bee and young larvae for 3 days. Its chemical contents and physiological functions are altered in accordance with storage conditions. Consequently, the quality control of RJ has gained much more attention in recent years as a commercial product. Although several countries have set the criteria for RJ, including China, Argentina, Bulgaria, and Japan, the international standards for RJ freshness have not yet been established. To access the fressness of RJ under different storage condition, we performed experiments to explore the dynamic variation of RJ proteins at -20 °C, 4 °C, RT, or 37 °C for 6 months. We found that proteins were degrading with the increasing of both storage temperature and time. And 45 proteins, among 286 proteins identified by LC-MS/MS, had significantly variations in abundance level, corresponding to the control sample (store at -80 °C). As to the abudance, the contents of major royal jelly protein 2 (MRJP2), MRJP3, MRJP4, MRJP5 varied in 4 °C and RT with the extension of storage time, especially in high temperature 37 °C for only three days, which were verified using monoclonal antibodies of MRJPs by Western-Blotting. Moreover, MRJP4 was more sensitive to storage condition. It degraded near a half when the fresh RJ stored in 4 °C for 4 w, RT for 2 w, or 37 °C for only one day. Hence, a couple of monoclonal antibodies of MRJP4 were used to produce the ELISA kit and immune-colloidal gold for the detection of MRJP4 in RJ. The ELISA kit quantitied the concentration the MRJP4 with the detection limit of 1.563-100 ng/mL, and the sensitivity of 3.758 ng/mL. And the immune-colloidal gold could detect the MRJP4 in fresh RJ with 1000 dilution in 10 min. Our results give a support to suggest a standard for RJ, and in view of proposing MRJP4 as an index for RJ freshness.

Characterization of Taiwan longan honeys based on phenolic compounds and flavor fingerprint characteristics

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National Ilan University, Ilan, TAIWAN

Longan honey is the most popular types of honey in Taiwan because of its distinctive flavor, especially high-priced Taiwan longan honey. However, the problem of honey adulteration has been a big concern. There are several different types of honey adulteration, the most difficult to resolve is the falsification of the floral or geographical origin of the honey. Therefore, the distinct authentication of its origin is great importance. In the present study, we collected 90 Taiwan longan honeys and used GC-IMS and LC/MS/MS to establish the multi-oriented fingerprint characteristics of these samples, including flavor characteristics and phenolic compound composition. 17 Thai longan honeys were compared . Overall, 15 phenolic compounds were determined in 107 samples by LC/MS/MS. Taiwan longan honey showed high 4-hydroxybenzoic acid, kaempferol, rutin and abscisic acid intensities and allowed the accurate classification of the honey samples according to origin by various techniques (including principal component analysis, cluster analysis and discriminant analysis). The flavor fingerprints of volatile organic compound (VOC) of honey samples were obtained from Gas Chromatography-Ion Mobility Spectrometry (GC-IMS). The results showed very good discrimination between honeys of two different geographical origins by PCA-LDA. Furthermore, the fingerprints provide superior resolving power for non-targeted profiling of VOC from highly complex samples such as honey.

Re-crystallization of honey – causes and prevention

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Intertek Food Services Gmbh, Bremen, GERMANY

Honeys generally tend to crystallize. Except for honey types which are usually present in a crystalline state, like canola or sunflower honeys, this effect is not desirable for liquid honeys. It leads to a hampering of further technological processing and to a turbidity of the product, which causes lower customer acceptance. Furthermore, a separation into a crystalline and a liquid layer reduces the microbial stability of the honey due to an accumulation of moisture in the upper section.
The period when honey begins to crystallize depends on several factors. As well known, it is its composition, particularly ratios between

specific carbohydrates and water, and furthermore the storage conditions. But other very important factors are the procedures of homogenizing and bottling the honey as well as the quality of packaging material.
In this presentation all these parameters will be described more precisely and illustrated with examples. All steps of conventional honey processing are screened with the focus to avoid a fast re-crystallization.
Beyond that a newly developed instrument will be introduced, by which it is possible to forecast a honey crystallization in an early stage. It will be shown how to use this device in order to improve the quality control of liquid honeys.

The investigation of antioxidant properties of propolis products obtained from Turkey

A. Tanugur Samanci [1], T. Samanci [1], E. Ozdemir [1], E. Capanoglu Guven [2]
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Propolis is a resinous bee product which has been collected by bees from the leaves and buds of trees such as poplar, eucalyptus, alder, pine, chestnut etc. and plants. The flavonoids and phenolic acids in propolis play significant role in its biological activity. The richness of propolis in terms of these constituents gives it a strong antioxidant property.
In this study, dry matter, total phenolic, flavanoid contents, total antioxidant capacities (CUPRAC method) and phenolic profiles of totally 30 propolis products sold in the Turkish market as drop forms were investigated.
Total phenolic, flavonoid contents, total antioxidant capacities (CUPRAC method) of the samples were analyzed with spectrophotometer. Their phenolic profiles were investigated via HPLC. The samples include different extracts of propolis whose solvents were classified as ethanol + water, water, glycerin + water, propylene glycol + water, glycerin and olive oil.
It was found that the total phenolic and flavonoid contents of ethanolic extracts of propolis were higher than those of aqueous extracts.
As a result of phenolic profile assay, the most frequently seen compounds were found as caffeic acid, cinnamic acid and ferulic acid. The number of phenolic acids and flavonoids detected in ethanolic extracts of propolis was found higher than those detected in aqueous extracts of propolis.
To sum up, it can be concluded that the solvent used in the extraction of propolis is an significant parameter affecting the antioxidant properties of the final product. This research showed that the number of studies investigating different parameters which affect the antioxidant properties of propolis should be increased. The findings to be obtained may elucidate the further studies regarding the standardization of propolis.

SYMPOSIUM 13:00-15:00
QUEEN REARING & INSTRUMENTAL INSEMINATION / BREEDING TECHNOLOGY I ROOM 517C

[Lead-off] Factors reducing losses of instrumentally inseminated honey bee queens in large scale production

M. Bienkowska
Research Institute of Horticulture, Division of Apiculture, Pulawy, POLAND

Honey bees are unique, in that selective breeding is based upon behavioral traits of a super organism with a complex social structure, in a constant state of change. Varroa mite and its associated pathogens are also undergoing constant change. For these reasons, honey bee breeding is challenging. Controlled mating is essential to achieve the goals of breeding programs, although difficult to control because bee queens mate in flight with several drones (10 to 20) in congregating areas. In most countries with a high density of colonies as well as a diversity of breeds and lines of bees in the apiaries, it is impossible to organize functioning natural mating stations. The only solution that guarantees controlled mating and in addition is independent of weather conditions is the use of artificial insemination. Poland is a country where bee queens insemination is well known and used routinely in bee breeding apiaries as well as for commercial production. Instrumental insemination with the semen of special breeds and isolated drones guarantees mating and selection purity. In the large scale queen production, it is important to determine the factors affecting the quality of these queens. In spite of many tests there are still problems with the onset of oviposition and losses of queens (up to 13% in Poland). The main causes of the loses are: queens not accepted by bees, non starting of oviposition, premature supersedure or careless treatment by beekeepers. The most important factor is semen residue in the oviducts, as this may prevent the start of egg laying and consequently death of the queen. Therefore, it is important to identify factors affecting quality, fitness and acceptance of instrumentally inseminated honey bee queens

Practical use of cryopreserved honey bee germplasm: benefits and limitations

B. Hopkins, S.W. Cobey, W.S. Sheppard
Washington State University, Pullman, USA

Apis mellifera is a highly diverse species comprised of 26 recognized subspecies, numerous ecotypes, and specialized strains. They have evolved a suite of different characteristics that has allowed them to survive in tropical and temperate climates in almost every region where they have been introduced. The diversity in the species is under threat for a number of reasons including human mediated movement within areas of endemism, pests and diseases and land management. All of these factors warrant a concerted effort to conserve honey

bee genetic resources for both conservation and breeding.
Cryopreservation of germplasm has the potential to serve both biodiversity and breeding goals. This presentation will cover ongoing honey bee germplasm collection and cryopreservation efforts by Washington State University. While we continue to work to improve cryopreservation methodologies, we report here the practical ongoing use of cryopreserved honey bee semen for breeding purposes. Cryopreserved semen has been used to produce queen mothers and breeder queen daughters that have been distributed to large-scale queen producers in the US. In addition, semen cryopreserved for up to 7 years has been used in the breeding of Caucasian honey bees (A. m. caucasica) that we have reintroduced into the US. The project has been ongoing for 8 years, resulting in the cryopreservation of more than 7000 microliters of semen representing germplasm from 4 Old World subspecies, 3 specialized breeding lines, and commercial US stock. Cryopreserved honey bee semen is now included in the National Animal Germplasm Program of the USDA.

Progress with the conservational biobanking of honeybee genetic resources

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In view of the continuing loss of honeybee biodiversity, the establishment of ex situ-reserves becomes an increasingly pressing issue. Here we present recent technical progress with the cryopreservation of drone semen, achieved in our laboratory, and present a biobank for honeybee resources that is currently being created in Germany.
In order to facilitate the testing of new cryoprotocols, we have compared 8 laboratory bioassays for semen quality regarding their capacity to predict fertility, and found that the conventional indicators of motility and viability were best suited for this purpose. Next, we evaluated the effect of hen egg yolk, a frequent additive to freezing extenders, on the and fertility of queens receiving unfrozen semen, and found no evidence for negative impacts. In a series of experiments, we found most common cryoprotectants to be toxic to queens/semen at the concentrations required for cryopreservation, including the frequently used dimethyl sufoxide (DMSO). We therefore developed a protocol to wash semen for the removal of cryoprotectants before insemination. Finally, we developed an innovative cryoprotocol for drone semen that involves the gradual addition of cryoprotectant through dialysis. It has been shown to yield between 3 and 98% female brood in the offspring of inseminated queens in a series of experiments, with numbers of sperm in spermathecae ranging from 20.000 to 3.8 million. Because inbreeding is likely to become a concern when small populations are to be maintained with the help of cryopreserved resources, we also tested the effect of low to moderate levels of inbreeding on semen freezability. No negative impact could be observed.
Together with the Bee Institute of Kirchhain (Germany), we (LIB) are now using the technological progress made by us and others to build up a conservational cryo-reserve of resources of A. m. mellifera and A. m. carnica, as part of the German National Cryobank for Husbandry Animals. Donors from other countries are offered the mirroring of resources in their own cryobanks. We envisage an initial collection of resources from 300 colonies by 2022, including not only semen, but also whole-body samples as well as purified DNA.

The Saskatraz Breeding and Selection Program 2019

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The Saskatraz® project was established in 2005 with the objective of breeding productive, gentle honey bees with tolerance to mites and brood diseases. Colonies showing excellent honey production, longevity and freedom from brood diseases on the Canadian prairies were initially close population mated to Russian stock and German Carnica lines. Colonies from these crosses selected for honey production and colony health were isolated at an apiary called Saskatraz®, and subjected to natural selection (cf www.saskatraz.com for review). This report describes the results of selecting for both honey production and varroa tolerance with no synthetic chemical miticides, from this diverse gene pool over a period of more than 12 years. The first six Saskatraz® families were selected after 3.5 years of natural selection. Since then stock from Buckfast lines, Carniolian and VSH phenotypes have been back-crossed to Saskatraz® families (17) and the progeny subjected to natural selection. Close population mating at natural selection apiaries of selected Saskatraz® phenotypes, with drone populations under high varroa mite infestations (survival colonies) is continually performed to enrich for varroa tolerance by recurrent natural selection. Methods are described for maintaining the selected lines by outcrossing, and recurrent selection to maintain and improve honey production by close population mating to diverse drone mother colonies showing excellent honey production and survival.
Extensive gene expression analyses of survival colonies from the Saskatraz® program has resulted in the identification of potential biomarkers to assist breeding programs in identifying varroa tolerant phenotypes. In a genome wide DNA microarray analyses of two extreme phenotypes for tolerance and susceptibility to varroa parasitism 200 differentially expressed genes were identified. Three of these genes (AmCbE E4, AmApoD and AmCYP6A1) showed increased expression in varroa tolerant phenotypes, after screening sixteen phenotypes with varying degrees of varroa susceptibility. We have also performed substantial kinome profiling of survival colonies with varying degrees of susceptibility to varroa mite parasitism. A set of kinome biomarkers was identified within individual bees which are predicative of a colonies susceptibility to varroa mites.

Quality and Performance of Imported and Domestic Queens

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The quality of a honey bee queen and her mating plays a pivotal role in determining the success and productivity of the colony. The goal of this project was to examine the variability among imported and domestic stocks in measures of initial queen quality, relate these measurements to colony productivity, and provide data to support management decisions.

We purchased queens from British Columbia (BC, n=43), New Zealand (NZ, n=42), and Hawaii (HI, n=43), analysed a subsample for morphometric traits, sperm count and viability, and introduced the remainder to colonies at three sites across Alberta, Canada, where they were evaluated across two production seasons. We measured colony-level parameters including the “patchiness” of the brood pattern, hygienic behaviour, disease-tolerance, honey production, adult bee and sealed brood populations, as well as cluster size and colony weight before and after winter.

The queens received were well-mated (mean 5.9 million ± 0.22 million sperm, range 3.1-10.4 million) stored in the spermathecae of an initial subsample of queens. There were, however, differences among sources for both queen weight and sperm count (NZ 5.3 ± 0.37 million, BC 6.48 ± 0.44 million, HI 5.93 ± 0.27 million). Sperm viability was also high across the stocks (mean = 85.71 ± 1.03%, min = 69.13%, max = 98.17%; NZ = 89.22± 1.80%, HI = 83.30 ± 1.58%, BC = 84.54 ± 1.66%). We also measured the sperm count of surviving queens sampled at the end of the experiment (NZ 4.0 million, n=10; BC 5.4 million, n=7; Hi 3.4 million, n=9).

We found significant differences among stocks. NZ queen-led colonies had less solid brood patterns and smaller brood and adult bee populations, despite the queens having initially good viability and sperm count; this was likely associated with the higher incidence of chalkbrood observed in these colonies, especially in 2018. We will discuss these results and other performance measures that varied among the stocks and sites. The majority of the queens we tested from commercial stocks did not survive through two production seasons, whether through lack of acceptance, supersedure or death.

Honeybee mating and its impact on queen performance and health

B. Baer
Center for Integrative Bee Research (CIBER), Department of Entomology, University of California Riverside, Riverside, USA

The mating biology of honeybees and other social insects is truly spectacular. Partner choice and copulations only occur during a very brief period early in the life of queens to acquire and store a life time support of sperm. Despite the fact that queens produce up to 1.7 million fertilized eggs, they never replenish sperm and therefore have to use sperm in a highly economic way. The initial processes occurring during mate choice, copulation, sperm competition and female choice as well as sperm storage are increasingly recognized as key determinants of queen performance and colony success. I will summarize the progress of work that has been conducted over recent years to understand the impact of mating on the life history of queens and provide several examples of how the use of proteomic and metabolomic analyses of sperm and reproductive secretions provide detailed insights into the processes that accompany sperm transfer and storage. Such knowledge allows to make accurate predictions about the processes present and their consequences, which can then be experimentally tested in the field. Such knowledge is also of value for future bee keeping, because it can be used for the development of novel tools of breeding and / or monitoring of bee health.

Drone factors influencing queen reproduction and health

E. Nino [1], L. Brutscher [1], B. Baer [2]
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Honey bees are major pollinators of agricultural and non-agricultural landscapes. High losses of honey bee colonies are being reported across the world and beekeepers frequently report poor queen quality and queen failure as one of the primary causes. Honey bee colonies are highly vulnerable to compromised queen fertility, as each hive is headed by one reproductive queen. Queens mate with multiple drones during a single mating period early in life in which they obtain enough spermatozoa to fertilize their eggs for the rest of their reproductive lifespan. The process of mating initiates numerous behavioral, physiological, and molecular changes that shape the fertility of the queen and her influence on the colony. For example, receipt of drone semen can modulate queen ovary activation, pheromone production, and subsequent worker behavior and physiology. These changes, if suboptimal, can lead to colony failure. In addition, seminal fluid is a major component of semen that is primarily derived from drone accessory glands. It contains a complex mixture of proteins such as proteases, antioxidants, and antimicrobial proteins. Seminal fluid proteins are essential for inducing post-mating changes in other insects and thus they may also impact honey bee queen fertility and health. However, the specific molecules in semen and seminal fluid that initiate specific post-mating changes in queens are still unidentified. Herein, we summarize the mating biology of honey bees, the changes queens undergo during and after copulation, and the role of drone semen and seminal fluid in post-mating changes in queens and potential subsequent benefits or consequences. We will also review potential roles for honey bee drone seminal fluid proteins in queen reproduction and health.

Further research elucidating the role of drone fertility in queen reproductive health has great potential to contribute towards reducing colony losses and advancing honey bee stock development as a sustainable way to overcome various stressors honey bees encounter.

SYMPOSIUM 15:30-17:30

QUEEN REARING & INSTRUMENTAL INSEMINATION / BREEDING TECHNOLOGY II ROOM 517C

Optimizing drone raising and marking techniques in Belgium: a report

S. Egyptien, F. Brutinel, J. Ponthier, S. Deleuze
Department of Veterinary Clinical Sciences, Companion Animals And Equids, University of Liège, Liège, BELGIUM

Research on drone semen freezing requires very large numbers of drones, which can be very challenging. No established technique to supply these large numbers has been clearly described. We report our attempts to reach sufficient numbers of drones while maintaining a viable balance in the breeding system under Belgian beekeeping conditions.
Controlling the age of drones can also be an issue. We report marking drones every 24h for up to 16 days using one different color POSCA® Marker per day. Colored drones were well tolerated by the workers.
Drones frames were introduced in a strong colony in a Dadant 10 frames hive for the queen to lay eggs.
24h before first hatching the frames were placed in a Dadant 6 frames hive and 2 different caging techniques were tested.
Technique 1: Males were kept on a frame in a cage that was opened for daily marking. We observed that the cage quickly got overcrowded and drones tended to escape or get crushed and killed during the manipulations. The technique was then slightly modified and drones were individually collected from the frame, marked and then placed on a caged workers frame. This, however, was associated with too a high number of drones flying away during the manipulation and was abandoned.
Technique 2: A maximum of 3 frames of males, 1 of workers and 1 of food were transferred into the body of a Dadant 6 frames hive placed on top of an empty super with a queen excluder between them. Date of birth was assessed by daily marking of the emerged drones. Drones were at first well tolerated but after one month they were chased out of the hive and killed by the workers. Drones raised in the small hive were smaller than drones that escaped and were raised by the nearby colonies. They also had diarrhea and almost no semen could consequently be collected.
We conclude that raising large numbers of drones in surrogate hives is suboptimal. An alternative where drones are raised in their home hive and kept in small groups in small cages should be investigated.

Impacts of imported honey bee germplasm on US honey bee populations

W.S. Sheppard, B.K. Hopkins, M.A. Taylor, S.W. Cobey
Washington State University, Pullman, WA, USA

The main importation of honey bees from Europe and north Africa to the United States took place between 1620 and 1922. In 1922, regulations restricted further importation and access to European honey bee stocks for bee breeding purposes was largely eliminated. Substantial and ongoing losses of managed and feral honey bees in the US over the past decade increased bee breeder interest to access genetic diversity from Old World source populations and subspecies. Using a permit to import honey bee semen, the bee breeding program at Washington State University has used both fresh and cryopreserved honey bee semen and instrumental insemination since 2010 to produce breeder queens of Carniolan and Caucasian background and to augment existing Italian stocks. Recently, we used microsatellite marker evidence to evaluate genetic diversity in honey bee populations belonging to commercial queen producers in the western US. Significant increases in genetic diversity were found in honey bee populations belonging to queen producers who incorporated imported germplasm compared to queen producers that did not. Old World honey bee populations represent a reservoir of genetic diversity for honey bee selective breeding efforts throughout its introduced range.

Queen reproductive quality and colony health in honey bees

D. Tarpy
North Carolina State University, Raleigh, USA

Survey results from beekeeping operations in the U.S. have consistently shown that one of the primary perceived problem for beekeepers is ‘poor queens.’ While this factor encompasses many different symptoms, most of these reports document premature supersedure (queen replacement), inconsistent brood patterns, early drone laying (indicative of sperm depletion), and failed requeening as indicative of low queen quality. Determining the factors that result in low-quality queens is therefore of fundamental importance for improving colony productivity and fitness. Our research has explored the connections among queen rearing, insemination success, management practices, and various measures of colony productivity and health that has helped to elucidate the central role that queens play in modern bee management.

Sperm viability assessment in honey bee drones using the OpenCASA system

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BIOFITER research group, IUCA, University of Zaragoza, Huesca, SPAIN

The quality of sperm produced by drones is essential to the reproductive success of the queen and may determine the colony's survival and level of productivity, as well as the success of instrumental insemination. In this context, the study of drone sperm quality is of great interest, both in basic and applied studies. Sperm plasma membrane integrity, which is also known as sperm viability, has been one of the most used parameters for sperm quality assessment in drones. In previous works, we developed a simple method to analyze sperm membrane integrity in mammals using an acridine orange-propidium iodide combination. We have also recently published the OpenCASA, an open-source software that allows the automatic analysis of mammalian sperm viability using fluorescence images. The aim of the present work is to adapt these methods to the honey bee for a faster and more precise determination of sperm viability in this species. A total of 21 mature drones, 7 males from 3 colonies, were sampled individually. Semen was diluted in Kiev buffer before evaluation. Sperm viability was determined using acridine orange and propidium iodide, after adjusting the concentration of fluorochromes to this species. At least 200 cells were examined per sample, both subjectively and using the OpenCASA software, and the results were compared using Pearson correlation coefficients. An average of 78.90 % and 76.99 % viable spermatozoa were obtained using manual and automated methods, respectively, and the results were highly correlated ($r = 0.991$). This study confirms that the use of acridine orange and propidium iodide combination, followed by the analysis of the images with the OpenCASA software is an efficient and precise alternative to sperm viability assessment in honey bee drones. This work was supported by the Spanish MINECO (grant AGL2017-85030-R), and the DGA-FSE (grant A07_17R).

Impact of Nutrition on the Queen's mating

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MIHE (Mirdamad Institute of Higher Education), Department of IT, Gorgan, IRAN

It is possible, with proper methodology, to reduce the queen loss during mating to less than 5 percent. Improving several conditions will reduce the mating losses. First, the nutrition of the nucleus colony must be high. Second, mating must take place during the hot season. Third, there must be large numbers of drones present. Four days after emergence the queen is mature. 90 percent of queens will mate between 7 and 10 days after emergence. In the hot season 90 percent of queens mate between 6 and 8 days after emergence. After 22 days the queen will no longer fly for mating. The first eggs are laid between 8 and 18 days after the queen's emergence. In a normal season 90 percent of the queens lay their first eggs between 12 and 14 days after emergence. In a hot season 90 percent of the queens lay their first eggs between 7 and 9 days after emergence. In our new method the nucleus box that contains a virgin queen is fed 100 grams of a mixture of honey, pollen, water, sugar, royal jelly and lemon juice. The feeding must take place on sunny, hot days, 7 to 10 days after the queen's emergence. With this method 90 percent of the virgin queens will fly twice for mating in a three-day period and the sign of the last mating will be present after the second flight. The time of day for the feeding is important. Queens will fly between 12:00 to 16:00 with 90 percent of the flights taking place between 14:00 and 16:00, therefore, it is important to feed the colony between 13:00 to 14:30.

Mating Measures: Comparison of honey bee subspecies mating behavior utilizing Radio Frequency Identification (RFID)

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The improvement of honey bee stocks through selective breeding represents a sustainable approach to assure future pollination services for food production and security. Inadequate mating can affect the capacity of queen producers to provide quality stock. This can be as a result of inclement weather, compromised environments and additional stressors. The goal of this research is to improve our understanding of mating flight behavior of various honey bee (*Apis mellifera*) subspecies utilizing Radio Frequency Identification Technology (RFID). In this study, we attached RFID tags to queen honey bees and drones to measure aspects of their orientation and mating flights. -
This research compared the mating flights of queens of *Apis mellifera ligustica* and *Apis mellifera caucasica*. These subspecies derive from two different evolutionary lineages endemic to regions having different climatic conditions. Virgin queens were reared according to standard commercial methods. Both initiation (timing) and duration of mating flights and drone flight behavior were recorded. Results from this research may assist queen producer efforts to optimize mating schemes for honey bee strains of differing genetic backgrounds. This collaborative research with engineers, beekeepers, and researchers which utilized RFID to compare and evaluate mating behavior provides fundamental improvement in understanding honey bee mating behavior. Such information will allow queen producers to better select, integrate, and propagate appropriate honey bee stocks relative to seasonal weather conditions. This can enhance rearing protocol best practices, and encourage quality control for queen producers and their beekeeper-customers.

Genetic aspects of emergence weight, ovary weight and number of ovarioles in honey bee queens

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The honey bee queen plays an important role in growth, productivity, colony survival and, at the phenotypic side, some physical characteristics have been linked with honey production, via reproduction potential. In fact, a recent study has shown that the honey production in the colonies with selected queens by emergence weight was, on average, 26% higher than with unselected queens. However, why colonies with heavier queens at emergence produce more honey? Is it related to the number of ovarioles? Queen's emergence weight, ovary weight and ovariole number were recorded on 500 virgin queens, from 40 different families. Variance components for queen's emergence weight, ovary weight and ovariole number were estimated using a one-trait animal model, considering missing drone information. Heritability for emergence weight (0.46) and ovariole number (0.31) were moderated and, for ovary weight was high (0.62). All traits have selection potential, however genetic correlations must to be estimated in order to well understand how these traits are related. Further studies will help to investigate the genetic correlation among emergence weight, ovary weight, ovariole count and honey production, as well as to increase the number of records in the future.

Instrumental Insemination of Queen Bees, Factors Affecting Success Rates

S. Cobey
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Instrumental Insemination is an essential tool for honey bee breeding, research and conservation. The technique provides a method to control honey bee mating for breeding purposes and to provide novel crosses for research. Selection for honey bee stocks that express increasing resistance to parasites and pathogens as well as demonstrate high colony performance is a goal of beekeepers worldwide. The conservation of genetic diversity and the preservation of threatened subspecies and ecotypes is a recognized need. Improved techniques to cryopreserve bee semen for stock maintenance and stock recovery purposes, also increases the need for this tool.

Instrumentally Inseminated queens, IIQS, have the capability of heading productive, full size colonies. Their performance and longevity are essential to evaluate and propagate stocks for breeding purposes. Proficiency and a high rate of success are dependent upon proper procedures and specialized beekeeping skills, including attention to detail and proper sanitation. Many factors influence the success rate and colony performance of IIQs. The pre and post insemination care of queens and rearing of drones to maturity, in numbers and the desired genetic sources, is critical to success. Practices must strive to optimize nutrition and ensure the proper mating age, banking procedures, CO2 treatments, semen dosage and queen introduction methods. The effect of seasonal conditions, the prevalence of parasites, pathogens and chemical residues must also be considered. Given the increasing need and use of I.I., it is essential to recognize and optimize the various factors that influence the performance of IIQs.

APITHERAPY

09 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

APITHERAPY - FROM SCIENCE TO PRACTICE I ROOM 517D

[Lead-off] Bee Products – Nutritional Value, Physiological Vs. Pharmacological Effects

C. Mateescu
National Institute for R&D for Food Bioresources, Bucharest, ROMANIA

Background: With an increasing interest, bee products are used worldwide as raw products or in food supplements but due to their proved therapeutic effects, in some countries, they are used as medicines. It is nowadays generally understood and accepted that the quality of these products should be defined in terms of physiological or pharmaceutical properties of their authentic components.
The aim of this study is to discuss and understand the specificity of action and the reference doses of the specific active ingredients in the bee products that can be used either for their physiological or pharmaceutical effect.
State-of-the-art: Food supplements are defined as concentrated sources of nutrients or other substances with a nutritional or physiological effect that are marketed in “dosed” form. They are intended to correct nutritional deficiencies, or to support specific physiological functions. They cannot exert a pharmacological, immunological or metabolic action. Therefore their use is not intended to treat or prevent diseases in humans or to modify physiological functions.
A medicine has specific properties for treating or preventing a disease; it is used either to restore, correct or modify physiological functions exerting thus a pharmacological, immunological or metabolic action.
In vitro and in vivo studies as well as several clinical trials have shown that bee products can be used as both food (honey, pollen, bee bread), dietary supplements (honey, pollen, bee bread, propolis, royal jelly) but also as active substances in the pharmaceutical industry (medicinal traditional products, homepathic drugs (bee venom, whole bee bodies) etc. Conditioning, processing technologies, recommended dosages, target organs or systems make it possible to obtain a product with an expected effect (physiological vs. therapeutic) but many others details, will make the difference between a medicinal product and a food supplement.
Conclusions: Border compounds in bee products and their stadardized specific active compounds, will be presented in an attempt to define whether they could really be “medicinal” products or how much of propolis, royal jelly or their fractions, bee venom and its active coumpounds would make the difference between a supplement and a medicine, between traditional and scientific approach on apitherapy.

Treatment of Post Surgical & Trauma Induced Scar Tissue with Bee Venom Therapy & Apipuncture; A Clinical Case Study

F. Keller
The American Apitherapy Society Inc, Northport, USA

Scar treatment with Bee Venom Therapy & Apipuncture(Traditional Chinese Medicine and Apitherapy) in Clinical Practice. Bee venom therapy (BVT) is an effective scar treatment for new and older ones which can cause considerable local pain, numbness and tingling as well as referred pain to another part of the body. Bee venom can alleviate these conditions and restore sensory motor function to local & distal areas. The techniques primarily utilized; mini stings, micro stings & full stings along side an acupuncture needle. Objective: increase blood flow to the area, dissolve underlying tissue impingement, relieve pain & restore proper function. Frequency & duration of treatment depends of several factors including size, age and depth of scar tissue. The effects of BVT are remarkable as will be presented via before & after photos of individual cases. Bee venom contains hyaluronidase which reduces inflammation, promotes circulation and essentially dissolves scar tissue. Bee venom therapy (BVT) is an effective scar treatment with excellent, promising results. Cosmetically bee venom softens, flattens out, reduces & significantly fades most scars including keloids. Proper precaution & bee venom allergy testing essential.

Effect of *Apis mellifera*, *Melipona beecheii* and *Frieseomelitta nigra* honeys on the expression of virulence genes of Salmonella enterica Serovar Typhimurium

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Salmonellosis is caused by bacteria of Salmonella genus, which have an important significance worldwide, mainly in underdeveloped countries due to health conditions, especially in children. This disease is commonly transmitted by food of animal origin such as meat, eggs and chicken, making it a public health problem. According to the World Health Organization, it is estimated that Salmonellosis annually affects thousands of people, causing approximately one hundred thousand deaths per year. This disease is generated by the species Salmonella enterica, which have many different serotypes, one of them is S. Typhimurium, frequently isolated from patients with acute diarrhea. It has been found S. Typhimurium strains that are resistant to streptomycin, cephalosporin among other antibiotics. An alternative to use of antibiotics is honey, which has been utilized for treating bacterial diseases due to its antibacterial properties. Yucatán is the biggest producer of bee honey produced by *Apis mellifera*, in México. However other bee species that produce honey, like the stingless bees *Melipona beecheii* and Frieseomelitta nigra nigra, are being cultivated in Yucatán and their honeys have been considered as medicinal. The main goal of this study was to evaluate the effect of the three honeys on the growth and virulence genes expression of S. Typhimurium SL1344, using the analysis of expression in transcriptional fusions of virulence genes linked to reporter genes lacZ o cat. The results showed that concentrations of 5% v/v of Apis and Melipona honeys, and 3% v/v of Frieseomelitta honey were able to diminish the growth of S. Typhimurium. On the other hand, concentrations of 3% v/v of Apis and Melipona honeys, and 1% v/v of *Trigona* honey were able to decrease both the Salmonella motility and virulence genes for flagellum biosynthesis and the localized in the pathogenicity island 1(SPI-1).

Successful and safe use of royal jell with careful attention to its cross-reactivity to certain allergens

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Royal jelly (RJ), a natural product secreted by honey bees, has been widely used for dietary supplements, beverages, medicines, and cosmetics. Unfortunately, however, RJ sometimes causes allergic reactions at first contact in persons with a history of any allergies, suggesting a possibility that they have antibody cross-reactivity against RJ. In the present study, we tried to investigate antigenic cross-reactivity between RJ and typical allergens with sera from atopic dermatitis (AD) patients.
Sera were collected from 30 AD patients (16 males and 14 females) who had never taken RJ. Their total IgE and specific IgE against 39 allergens were evaluated in an outside laboratory, and IgE antibody titers to RJ were measured by ELISA, which has been established in our laboratory. Then, antigenic cross-reactivity of RJ with common allergens was investigated by ELISA-inhibition assay using RJ-reactive sera.
As a result, interestingly, sera from 10 out of 30 AD patients showed a positive response to RJ. Correlation analysis revealed that RJ-antibody titers had significant relationships with non-specific IgE levels and the allergic scores for arthropods, such as crab, shrimp, mite, and cockroach. The subsequent ELISA-inhibition assay clearly demonstrated that European house dust mite (Dermatophagoides pteronyssinus) is, at least, one of the cross-reactive antigens of RJ. The present results may explain unforeseen allergic reactions in persons with any allergies, who have specific cross-reactive antibodies to RJ. Therefore, further studies are needed to clarify the relationship between RJ and other major allergens such as crustaceans for the successful and safe use of RJ.

Royal jelly has beneficial effects on lipid profile, satiety, inflammation and antioxidant capacity in asymptomatic overweight adults

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Obesity and overweight are chronic disorders of multifactorial origin that are characterized by high oxidative status and by chronic activation of macrophages in peripheral tissues. Effective therapeutic approaches to lower inflammation and oxidative stress are currently of general interest. Royal jelly (RJ) is a functional food with a broad range of pharmacological activities, manly used by healthy individuals or borderline patients to protect themselves against disease onset.
The objective of this randomized, double-blind, placebo-controlled trial was to investigate the effects of RJ supplementation on oxidative and inflammatory parameters in asymptomatic overweight adults, considered at an early stage of developing metabolic syndrome. The experimental group (N = 30) was given RJ (2000 mg daily) and the control group (N = 30) was provided with a placebo for eight weeks. Anthropometric, biochemical parameters, and biomarkers of oxidative stress and inflammation were assessed at baseline, after 4 and 8 weeks of the intervention, and after additional 2 weeks of follow up.
Compared with the placebo, RJ supplementation demonstrated a statistically significant decrease in total cholesterol (-6.7%, $p = 0.041$) and inflammatory marker C-reactive protein (-19%, $p = 0.027$), whereas significant increases were observed in anti-inflammatory marker adiponectin (+34%, $p = 0.011$), endogenous antioxidants bilirubin (+35%, $p = 0.002$) and uric acid +(5%, $p = 0.018$), total antioxidant capacity in serum (+54%, $p = 0.005$), and serum leptin levels (+17%, $p = 0.025$).
The present results support RJ's antioxidant, anti-inflammatory and hypolipidemic activities, previously studied mostly in animal models and in selected groups of patients.

Colombian propolis with biological potential: antitumor and immunomodulatory action, in vitro assay in osteosarcoma cells

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Propolis is a product collected and processed by bees of the species *Apis mellifera* and has a wide spectrum of potent biological activities. The main chemical components of propolis are flavonoids. Flavonoids have diverse therapeutic effects such as arrest of cell cycle,

induction of apoptosis and modulation of antitumor immunity. For this reason, propolis has been postulated as a compound with cytotoxic action in tumoral cells. Some of the molecular mechanism involved in the genesis of cancer include disruption at checkpoints of the cell cycle and activation of external signals that promote rapid cell division. In this study, we investigated the expression of two genes: p21 and p53. p21 encodes the CDKN1A protein and blocks the CDK2 and CDK4 complexes avoiding the progression of the cell cycle in the G1/S phase in response to increased transcriptional activity of the tumor suppressor gene p53. Additionally, the role of interleukin-6 (IL-6) that may promote an immune response modulated by T-cells with antitumor activity.
Therefore, the objective of the study was to evaluate the effect of colombian propolis Ethanolic Extract (SIL) (25 ug/mL) in the relative expression of genes: p21 and IL-6 in an in vitro model of canine osteosarcoma (OSCA-8) after 48 hours of exposure and correlate it with previous findings of cytotoxicity. The gene expression was assessed by using the quantitative PCR technique (qPCR). There was a significant increase in the expression of genes p21 and IL-6, higher than the one caused by doxorubicin (chemotherapeutic agent). The overexpression of the p21 gene in the OSCA-8 cells could suggest that this would be one of the targets of the SIL extract that would help to explain its antiproliferative effect, as well as the IL-6 that might be favoring the signaling mediated by immunity adaptive antitumor.
In summary, this in vitro exploratory study in osteosarcoma cells provides some insights to understand the molecular mechanisms of action and targets of propolis SIL involved in cytotoxicity, and also provides a new approach for colombian propolis SIL that could be applied in the future in osteosarcoma therapy.

Infant's milk fortification with natural honey improves some hematological parameters and enhances body weight gain

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National Center For Research, Environment & Natural Resources And Desertification Research Institute, Khartoum, SUDAN

The issue of providing adequate foods for infants is quite sensitive and drags the attention of workers in the field of nutrition, as giving suitable supportive diet to the infants is considered one of the very important pillars of health particularly when honey is being addressed. This study was conducted in model medical center in Khartoum state of Sudan with objectives, to study the impact of feeding honey-fortified milk for 1½ - 2 years old infants in their weight gain and health as well as to assess the nutritional quality of honey. The targeted population consisted of 22 infants divided into 3 groups. Whereas group A & B consisted each of, 9 infants fed with 2 & 4 % honey fortified milk; respectively and group C consisted of 4 infants was considered as control and fed with plain milk. Infants were initially weighed and blood samples were taken for further hematological analysis to test total red blood cells count (TRBC), total white blood cells count (TWBC) and hemoglobin (Hb %). The obtain measurements were primarily manipulated with Microsoft office excel 2007 and then transferred to IBM SPSS Statistical program version 19 for further analyses of means, standard deviations, graphs, and analysis of variance. The results showed that feeding infants with honey- fortified milk significantly ($P < 0.05$) increased the infant's weights by 0.63 and 0.7 kg ;respectively for group A & B compared to group C. Also blood parameters were improved significantly. TRBC increased by 0.3 x 1012 and 0.4 x 1012 respectively for group A & B compared to group C. TWBC decreased by 1328.5 & 1096.3 respectively for group B & A compared to group C. Hemoglobin was increased by 5 % in group A and B when compared to group C. The study concluded that fortifying infant's milk with natural honey has positive health rewards.

SYMPOSIUM 13:00-15:00
APITHERAPY - FROM SCIENCE TO PRACTICE II ROOM 517D

[Lead-off] Use of Honey and Propolis in the Prevention and Treatment of Wounds

S. Stangaciu
Romanian Apitherapy Society, Mereni Contesti (DB), ROMANIA

Introduction: We asked ourselves what the common features are belonging to both honey and propolis that explains their so wide use, all over the world, since thousands of years, against so many kinds of wounds.
Finding these common features will help the practitioners to create better treatment protocols.
Aims: To offer these common features that respect the principles of Integrative Medicine to all practitioners and patients in need.
Materials and Methods: We have studied scientific and practical international literature on apitherapy. We have communicated with apitherapists from over 50 countries, in the last almost 30 years. We made a synthesis of our personal clinical experience.
Results: Both propolis and honey share a large group of substances that belong mainly to the groups of polyphenols and minerals which explains their over 10 similar effects. When used together with a healthy diet and lifestyle, honey and propolis can promote highly useful synergistic effects that explains the fast improvement or cure of most of the wound's types.
Conclusion: In the present research work, we show that after a detailed diagnosis of all main causes of the causes of the wounds, after a clear change in the diet and the lifestyle, by using well adapted, individualized formulas of honey and propolis extracts, both locally (topically) and systemically (orally), good and very good results can be obtained in a shorter period of time than by using classical, antibiotics and cortisone based drugs.

Antimicrobial effects of Bee Venom (BV), Propolis and N-Chromosome Royal Jelly (NCRJ) on antimicrobial resistant gram-positive bacteria, Streptococcus Pyogenes and Enterobacter

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The rapid development of antimicrobial and multi-drug resistant pathogens have become a great global concern, causing a significant rate in morbidity, mortality, the need for surgical intervention, and increasing the length of hospital stay and overall cost of treating the infections. Streptococcus Pyogenes can cause a wide range of diseases from ear and sinus infections to pneumonia and bloodstream infections. There are at least 517,000 deaths globally each year due to severe S. Pyogenes infections; 15%-20% of school-aged children have S. Pyogenes - 111 million reported world-wide each year. Enterobacter has multiple disease-causing pathogens among them include eye and skin infections, meningitis, bacteremia, pneumonia, CNS (Central Nervous System infection), intra-abdominal infections, septic arthritis, brain abscess, intestinal and urinary tract infections. In infants the morality rate from enterobacter is 50% to 75%.
The aim of this study was to investigate the effects of antimicrobial properties of BV, Propolis and NCRJ on these pathogens. Several microbiology tests were performed in Iran over the period of 3 years, from 2017-19, through the streak-plate procedure, isolating pure cultures of bacteria. The results of antibiogram tests showed both Streptococcus Pyogenes and Enterobacter have sensitivity towards antimicrobial properties of these bee by-products. Further studies are still required in order to reach a firm conclusion.
In this study, BV, Propolis and NCRJ were used, topically and/or orally, on 79 patients, ranging from 2 years old to elderlies, according to the patients' needs and condition. The dosage was decided based on their immunity system. Above 62 people were completely treated over a period of one month and the remaining shown significant improvement.
In this presentation, the methods of production and application of BV, Propolis and NCRJ will be discussed in more details and the results of laboratory tests will be given.

Study of possible transmission of bacteria, clinically important for humans, through a hymenoptera sting

I. Iliadis, D. Graikini, E. Giaouris, A. Papachristoforou
University of the Aegean, Myrina, GREECE

It has been suggested that the sting of the honeybees can induce pathogenesis or can even be fatal for humans, as a result of the transmission of bacterial pathogens from the stinger to subjects' bodies. Though this hypothesis contrasts with research indicating the antimicrobial activity of bee venom, there are some rare cases in scientific literature assuming the transmittance of bacterial infections (i.e. from Streptococcus pyogenes) from honeybee sting could be the cause of death in humans.
Since bee venom and bee sting-acupuncture is extensively used in Apitherapy, we investigated the possibility of the transmission of some clinically important for human bacteria (i.e., Staphylococcus aureus, S. pyogenes, Enterococcus feacalis/faecium and Pseudomonas aeruginosa) via the sting or through simple contact with honeybees.
A total of 144 forager honeybees were collected from the apiary. Experiments were conducted in three different ways: 1) a simulation of honeybee stinging was performed in the laboratory with live honeybees stinging directly the nutrient substrates; 2) honeybees were forced to sting sterilized leather (human skin simulation) and immediately afterwards the sting apparatus was transferred to the nutrient substrates; 3) the back legs, that are always in contact with honeybees' victims during stinging incidents, were removed and were attached to the nutrient substrates. To isolate the targeted bacterial species, four different selective media were used: Baird Parker agar supplemented with egg-yolk tellurite for staphylococci; blood agar base supplemented with streptococcus selective supplement for streptococci; Kanamycin Aesculin Azide agar for enterococci; and Pseudomonas CFC agar for pseudomonas. Following inoculation, all media were aerobically incubated at 37C for 24h, except BP agar for which incubation was extended to 48h.
Despite the extended number of replications, no colonies of pathogens were recovered. This suggests that honeybees do not carry any of these specific hazardous microorganisms for humans here tested.
A second replication of the entire project as well as a similar test using wasps and hornets species have been applied and further results are expected soon.

The use of Honey as Apitherapy remedy in Veterinary Medicine

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In the last few years, the use of natural products for animal care has progressively increased, meeting the needs of owners that are becoming more health-conscious but also representing a choice towards the reduction of the use of conventional medicines.
Apitherapy, the therapeutical use of bee products, is a discipline with great potential for development, both in the human and veterinary field, that can be applied to almost all animal species.
Even if most of the studies have been based on human medicine, many others have used animals for testing purposes, making it possible to apply the results to pets and in animal husbandry.
Honey is the most known bee product and thanks to its innumerable properties has been subject of numerous researches.

Its composition is closely related to its botanical and climatic origin and linked to the species of bees from which it is obtained.
Known for its nutritional value, honey in recent times has also been introduced as pharmaceutical support and in clinical practice,thanks to its bioactive substances, many of which have been chemically analyzed and characterized.
Honey is very rich in polyphenols, a family of complex molecules, present in different concentrations depending on the type of honey. Among them, flavonoids and phenolic compounds are characterized by antioxidant properties capable of neutralizing the action of free radicals. Such property, combined with honey's specific chemical and physical characteristics, provides it with its typical healing actions.
In recent years there has been a growing interest towards antibiotic resistance that implies serious risks to public health, both for humans and animals. The European Commission has undertaken a strategy aiming at fighting against this threat by monitoring programs and the identification of alternative solutions to the use of antimicrobials. For this reason, the interest in products used in apitherapy may increase in the next few years, thanks to their certified healing properties and the absence of residues and resistance risks, stimulating further researches especially in the veterinary field.

From science to practice: the bee venom therapy

E.C. Nyonta
[1] *Ets John Carafa Retraite A Bangoua, Bangangte, CAMEROON*

the second report from science to practice: the bee venom therapy since the bee sting inoculates his venom directly in the blood flux from capillaries, all microbes that circulate in blood are immediately paralyzed; it is about salmonellas of the typhoid, microfilaria and the sexually transferable illnesses. i am one of these patients that saw his filarial disappeared. the case that astonished us is more the one of a 59 years woman that confided us that the 25 sittings of bee venom that she underwent in our branch of bafang, had made disappear her white losses. By using the venom of bee, we noticed that the young children who endured the meningitis, aftermaths of this illness improve progressively. this is how we can take the risk to conclude that if a patient of meningitis follows treatment of modern medicine and the venom of bee simultaneously, these aftermaths can disappear completely to his recovery.
There are as many research tracks as we can lead only with the support of apimondia.
1 - others cases in our center of bangoua we have 3 cases of encephalopathy
The 1st case concerns a girl of 20 years exit of a hospital in october 2017 with for diagnosis: polyarthritis stiffness (that means she can't stand up without a help). The 2nd and 3rd cases concern 2 boys aged of 2 and 3 exit of hospitals in june 2018 with for conclusion of the cerebral scanner: atrophy cortical. these children had this illness since they were born. Conclusion
you agree with me that the apipuncture is an alternative medicine that can be developed and modernized to take care efficiently of the poor populations of under developed countries. thus, apimondia has the responsibility and the duty to innovate in this domain to make people know better through the world over. i would like to invite you to bangoua where we are already advanced in the apitherapy with our 20 hives and we are going to save together many poor that stagnate in misery and illnesses in particularly in cameroon and in sub-saharan africa in general.

How to Use Acupuncture to Stop Suffocation During Anaphylactic Shock or Asthma Attacks

X. Zhou
Elegant Bee Clinic, Alhambra, USA

Suffocation is the leading cause of death for asthma attacks and anaphylaxis. This presentation reports the method regarding how to use acupuncture to stop suffocation during anaphylactic shock or asthma attacks. A new hypothesis regarding the mechanism of asphyxia caused by an asthma attack or anaphylaxis was postulated based on two death cases presented by professor Changsan Xin in a class and observation of human winter swimming. Formula M.H. from Treatise on Febrile Diseases Caused by Cold (Shanghan Lun) should be used in such circumstances according to Traditional Chinese Medicine. However, it was impossible to administer oral medicine to choking or suffocating patients. A Chinese needling method administered on the nose, which could achieve a similar effect as Formula M.H., was then chosen for this study. Patients with asthma attacks during clinic visits were used to verify this technique and new hypothesis. It was discovered that the selected needling method can relieve the symptoms of asthma attacks in less than 2 minutes, ranging from 15 seconds to 90 seconds. The same technique was then used on patients with signs of suffocation or airway constriction caused by bee sting treatments. The itchiness, throat discomfort, shortness of breath, or suffocation was completely relieved in less than 2 minutes. Moreover, how to differentiate the symptoms of this type of anaphylaxis from other types are explained in detail during the presentation. A temporal procedure to stop suffocation with the Chinese needling method is proposed for medical experts and public review.

SYMPOSIUM 10:00-12:00
VALIDATION OF APITHERAPY IN MODERN MEDICINE I ROOM 517D

[Lead-off] Apitherapy, how does it work: from mechanism to practice

B. Lyoussi
University Sidi Mohamed Ben Abdallah, Laboratory of Physiology-Pharmacology -Environmental health, Fez, MOROCCO

In earlier civilizations as well as in many cultures today, the medicinal value and health promoting properties of beehive products have been accepted. Indeed, presently, it is enjoying renaissance at a global level. Apitherapy is part of the traditional medicine and the science and art of maintaining health based on bee product use. Since, oxidation/peroxidation involving reactive oxygen species (ROS) is an important contributor to the cause of major chronic diseases, there is emphasis on determination of antioxidant activity of bee products, and their protection against ROS-induced damage in human diseases.
Extensive research has shown the therapeutic promise of the use of bee products in enhancing health values and providing evidence-based data demonstrating their benefits in animal models of diabetes, hypertension, nephrotoxicity, and hepatotoxicity. In vivo, in vitro studies of several biomarkers of disease and pharmacological targets reveal the synergistic interaction of the bioactive constituents of honey, propolis, pollen and beebread in favor of their overall protective, preventive, and therapeutic effects, principally as a result of their anti-oxidant/oxygen radical scavenging activity, mainly due to the presence of numerous types of phytochemicals, such as phenols at high concentration.
In this presentation, we will discuss the physiological basis for the use of honey, propolis, pollen, beebread and their combinations, their current clinical uses, as well as the potential role of bioactive compounds in suppressing oxidative processes, and their potential to treat and/or prevent urinary calculus, crystalluria and proteinuria, leading to their use in the management of liver and renal diseases by maintaining the activity of antioxidant defense system.
Several studies including our own research provide a potential mechanistic basis for therapeutic effects of bee products, which in part, are mediated by synergistic action of their bioactive components. Identifying their exact mechanism(s) of action will allow better understanding of their pharmacological properties and promote their wider translation into clinical practice.
In conclusion, these data will open up a new perspective for the development of synergistic potential of bee products in treating various pathologies and will promote apitherapy in clinical practice, opening up new scenarios in green drug discovery, as well.

Galangin and Pinocembrin from Propolis Ameliorate Insulin Resistance in HepG2 Cells

H. Zhang
Institute of Apicultural Research, Chinese Academy of Agricultural Sciences, Beijing, CHINA

Insulin resistance has a critical role in type 2 diabetes. The aim of this study was to investigate the effect of pinobanksin, galangin, chrysin and pinocembrin from propolis on insulin resistance. Our study shows that galangin and pinocembrin can ameliorate insulin resistance; on the contrary, pinobanksin and chrysin are ineffective. Galangin and pinocembrin treatments substantially increase glucose consumption and glycogen content by enhancing the activities of hexokinase and pyruvate kinase. Galangin treatment with 80 μM increased hexokinase and pyruvate kinase activities by 21.94 % and 29.12 %, respectively. Moreover, our results demonstrate that galangin and pinocembrin can have a synergistic effect on the improvement of insulin resistance via Akt/mTOR signaling pathway, through distinctly up-regulating the phosphorylation of IR, AKT, and GSK3β, and remarkably down-regulating the phosphorylation of IRS. Most notably, this is the first study to our knowledge to investigate pinocembrin about the alleviation of insulin resistance. Our results provide compelling evidence for the depth development of propolis products to ameliorate insulin resistance.

Trans-10-hydroxy-2-decenoic acid protects against LPS-induced neuroinflammation through FOXO1-mediated activation of autophagy

M. You, Y. Chen, J. Tian, F. Hu
College of Animal Sciences, Zhejiang University, Hangzhou, CHINA

Neuroinflammation is thought to be implicated in the pathogenesis of various neurodegenerative diseases. We have previously reported that royal jelly (RJ) has the anti-inflammatory effect on microglial BV-2 cells. However, components contribute to the anti-inflammatory effect of RJ were largely unexplored. In this study, we first observed that trans-10-hydroxy-2-decenoic acid (10-HDA), the exclusive lipid component in RJ, could protect against LPS-induced neuroinflammation and neuronal apoptosis both in vivo and in vitro. To determine the extent of global inflammatory changes after 10-HDA treatment, we performed an RNAseq transcriptomic analysis. Compared with LPS treated BV-2 cells, 10-HDA pre-treatment significantly decreased levels of multiple pro-inflammatory molecules via regulating TNF-α/NF-κB axis and NLRP3 inflammasome. We further illustrated that 10-HDA inhibited neuroinflammation by triggering cell autophagy, evidenced

by elevated level of microtubule-associated protein 1 light chain 3-II (LC3-II) and the degradation of SQSTM1. More importantly, 10-HDA increased the expression of FOXO1 at the mRNA and protein level as well as reversed LPS-induced FOXO1 nuclear localization. Inhibition of FOXO1 and autophagy markedly reversed the alleviating effect of 10-HDA on neuroinflammation, indicating that 10-HDA dampens the activation of TNF-α/NF-κB pathway and NLRP3 inflammasome in BV-2 cells by modulating FOXO1-mediated autophagy. Collectively, these data uncovered that 10-HDA may be an interesting candidate for clinical evaluation in the treatment of neuroinflammation-associated diseases.

Standartisation of bee venom therapies

J. Körmendy-Rácz
Omme - Hungarian Apitherapy Association, Budapest, HUNGARY

It is very important, that all complementary methods are comparable with the standard medical procedures. One patient has to know exact, which healing method what kind of advantages, disadvantages has, and what is the effectivity of the treatment.
For the apitherapy applications it is essential, that treatment protocols are evaluated on a clinical way to obtain exact, detailed and relevant data. China is pioneer on this huge work. They set the goal to take the steps on the standartisation.
As first we developed two protocols, and give it to the bid audience: low back pain and rheumatoid artritis. The protocol includes: include criteria, exclude criteria, follow up parameters and method, the treatment protocol and the additional suggestions, diary parameter.
With the time we hope to be able to collect enough data to evaluate the protocols, compare them, and give statistically relevant information form medical professionals, apitherapists. We kindly ask every practicioner to join our initiative.

Vascular cell adhesion gene expression is downregulated by colombian propolis in osteosarcoma cells

D.P. Pardo Mora [1], O.J. Murillo Torres [2], M. Rey Buitrago [2], L. Lopez Kleine [3], A.K. Rodriguez [4], O.A. Torres García [1], J.M. Sforcin [5]
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The information about the cytotoxic activity and chemical composition from Colombian propolis is limited. Therefore, a Colombian propolis sample chemically characterized rich in triterpenes and flavonoids was used to evaluate the cytotoxic effect on canine osteosarcoma (OSA) cells treated with propolis ethanolic extract. We identified differentially expressed genes to understand which pathways could be involved in propolis cytotoxicity. Lactate dehydrogenase (LDH) assay was used to evaluate cellular death. The effect of propolis solvent (ethanol) and doxorubicin (Dox) was also evaluated. The Canine Gene 1.0 ST array (Affymetrix) was used to determine the gene expression profile of treated OSA cells, OSCA-8. The microarray results were validated by quantitative real time PCR (qPCR).
LDH assay showed cytotoxic propolis effect in canine OSA cells, dependent of doses and incubation time. After 72 hours of treatment with propolis, the cytotoxic concentration 50% (CC50) in OSCA-8 cells was lower than 30 µg/mL. Enrichment and annotation analysis were applied to obtain differentially expressed genes. VCAM 1 - vascular cell adhesion molecule 1, NRXN1 - neurexin 1, EFEMP1 - EGF containing fibulin extracellular matrix protein 1, COL14A1 - collagen type XIV alpha 1 chain and SFRP2- secreted frizzled related protein 2 were downregulated ($p<0.01$), while MMP3 - matrix metallopeptidase 3, CXCL8 - C-X-C motif chemokine ligand 8, ABCB1 - ATP binding cassette subfamily B member 1, PTGS2 - prostaglandin-endoperoxide synthase 2 and TSPAN8 - tetraspanin 8 were upregulated ($p<0.01$). The genes are involved in several metabolic processes; however, VCAM 1 is a gene which product is the vascular cell adhesion protein 1 and was downregulated in OSA cells exposed at Dox and propolis. VCAM 1 gene is involved in a wide spectra of biologic processes such as hemostasis, immune and inflammatory response. Some studies have shown an increased expression of VCAM-1 in breast cancer cells suggesting that VCAM-1 is a potential target for molecular intervention in carcinogenesis (Wang et al, 2014). A downregulated VCAM expression indicated that pathways associated with its expression may be involved with the antitumor effect of propolis in OSA cells.

Apitherapy in external otitis of dogs and cats, clinicals observations

P. Garcia
Clinique Vétérinaire de la Crau, Saint Martin de Crau, FRANCE

Ear infections of dogs and cats are an extremely common reason for consultation.The many therapeutic failures with classically used treatments often lead to the selection of resistant germs.
Antibioresistance is a health priority requiring alternative treatments.
The clinical observations on more than a dozen cases we have been able to make suggest that hive products can be an interesting alternative.

Michigan Honeys Exhibit Superior BioActivity Level (BAL)

F. Ozturk [1], K. Ramon [2]
[1] *School of Science and Technology, San Antonio, TX, USA,* [2] *Alma College, Alma, MI, USA*

Throughout history, honey has been used for various therapeutic purposes, such as wound healing, treatment of skin and eye infections, gut diseases, as well as a painkiller due to its high biological activity. The bioactivity potential of honey is mostly dependent on its biological and chemical constituents. Honey contains over 200 compounds, being broadly composed of sugars, water, amino acids, vitamins, minerals, enzymes, phenolic acids, and flavonoids. Honey has been reported to have various biological activity properties, such as immunomodulatory, antibacterial, antifungal, antidiabetic, anti-inflammatory, and antitumor activities. The exact composition of honey, thus the bioactivity potential, differs depending on the diversity of geographical region, climate, honeybee strain, and nectar source.
In our study, we characterized the biological activity potential of 159 honey samples collected from different regions of Michigan through identification of their antioxidant and antimicrobial properties, as well as physical properties.
The mean moisture content, pH, and color characteristics of the honey samples were 17.50 ±0.015%, 3.42 ±0.31, and 29.6 ± 22.74mm Pfund, respectively. The mean total phenolic acids and FRAP values were 135.25 ± 132.85 GAE mg/kg and 429.01 ± 258.29µMFe(II)/100g and 36.95 ± 20.53%, respectively. The antimicrobial activity of Michigan honeys against the Staphylococcus aureus is ranged from 0.72% to 91.38%. Total bioactivity potentials of these honey samples were calculated based on a formulation considering their physical, antioxidant, and antimicrobial properties, and presented as BioActivity Level (BAL) units.
Among the different types of monofloral and multifloral honey samples analyzed in our study, buckwheat honey of Michigan exhibited the highest BAL value due to its superior antioxidant and antimicrobial properties. Overall, our study is the first large-scale analysis (n:159) of Michigan honey and demonstrates that some of the investigated honey samples are good sources of total phenolic acids with high antioxidant and antimicrobial properties, which deems further analysis to be classified as medical grade honey.

SYMPOSIUM 13:00-15:00
VALIDATION OF APITHERAPY IN MODERN MEDICINE II ROOM 517D

[Lead-off] Apitherapy in the Web of Science

A. Timucin Atayoglu [1], A. Guner Atayoglu [2]
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Apitherapy has been practiced since the ancient times therefore references to apitherapy can be found in traditional medicine practices. The issue of evidence-based validation of apitherapy brings together current thinking and practice in the areas of characterization and validation of bee products. According to modern scientific publications, bee products have been found to exhibit bioactivities, such as antimicrobial, anti-inflammatory and antioxidant activities. “Web of Science” is an online subscription-based scientific citation indexing service that provides a comprehensive citation search. It gives access to multiple databases which allows for in-depth exploration within an academic or scientific discipline. It has indexing coverage of scholarly books, peer reviewed journals, original research articles, reviews, editorials, chronologies, abstracts, as well as other items from the year 1900 to the present. The “impact factor” of an academic journal is a measure reflecting the yearly average number of citations to recent articles published in that journal. It is used as a proxy for the relative importance of a journal within its field. The use of impact factor as a measure of quality is widespread. This review analyzes the current situation of Apitherapy including the in vivo and in vitro potential of selective bee products based on the Web of Science.

Effect of Propolis on Inflammatory Diseases and Its Mechanism

X. Zhang
Anhui Liapin Bee Technology Co. Ltd., Hefei, CHINA

Propolis, a resinous substance with aromatic odor which collected by honeybee from plant buds,trunk and wound, and mixed with the secretions of the maxillary gland and the wax gland, which possesses a broad spectrum of pharmacological and biological activities. The good anti-inflammatory effect is one of the first been concerned pharmacological activities of propolis. This article reviews the anti-inflammatory active ingredients of propolis and its mechanism of action, providing theoretical basis for the clinical application of propolis in inflammatory diseases and adjuvant treatment of cancer, cardiovascular diseases, diabetes and other complex diseases.

Antimicrobial effects of Bee Venom (BV), Propolis and N-Chromosome Royal Jelly (NCRJ) on antimicrobial and multi-drug resistant gram-negative bacteria, Pseudomonas, Acinetobacter and E.coli

M. Moradi Chamachar [1], M. Mohammadi [1], H. Yeganehrad [2]
[1] *Department of Biology, Science And Research Branch, Azad University, Tehran, IRAN,* [2] *Caspian Apiaries, Delta, CANADA*

Pseudomonas and Acinetobacter pathogens cause an increasing number of healthcare associated infections with significant morbidity and mortality, which are often associated with ICU admission. Serious Pseudomonas can cause blood infection and pneumonia. Acinetobacter

causes bacteremia, pneumonia, meningitis, urinary tract infection, central venous catheter-related infection, and wound infection. Some E. coli can cause bloody diarrhea, urinary tract infections, severe anemia or kidney failure, which can lead to death.
The purpose of this study was to examine the antimicrobial properties of BV, Propolis and NCRJ and their possible effectiveness against these pathogens. Several microbiology tests were performed in Iran over the period of 3 years, from 2017-19, through the streak-plate procedure, isolating pure cultures of bacteria. The results of antibiogram tests showed Pseudomonas had no sensitivity, but Acinetobacter and E.coli have sensitivity towards antimicrobial properties of these bee by-products. Further studies are still required in order to reach a firm conclusion.
Another studies were done on 32 patients, above 15 years old, using BV, Propolis and NCRJ, topically and/or orally, according to the patients' needs and condition. The dosage was decided based on their health condition. The result for Pseudomonas was slightly different in compared to antibiogram tests; consuming NCRJ and Propolis increased the immunity system of the patients to fight the infection. NCRJ and Propolis can be used as an alternative to antibiotics.
In this presentation, the methods of production and application of BV, Propolis and NCRJ, as well as the results of laboratory tests will be discussed in more details.

Apitherapy in Veterinary Medicine: Best Clinical Cases by Medical Speciality

M.A. López Pazos
Veterinary Medical Doctor, Apitherapy & Chinesse Medicine Specialist, Temuco, CHILE

Based on the traditional and biomedical history of the bee products – using the properties of each product of the beehive and the synergy of them – I developed different Treatment Protocols for different pathologies. Far and above Apitherapy, the protocols combine many different therapeutic tools (Apitherapy, Traditional Chinesse Medicine, Phitotherapy, Nutrition Support, Allopathic Modern Medicine), to treat and support many animal species. However, Apitherapy has been determinant in therapeutic results of real clinical cases that I treat. Eighteen years of work on Clinical Integrative Veterinary Medicine show that the beehive products are fundamental elements on the outstanding results CENTROVETERA – Apitherapy Veterinary Medical Center – offers.
For best results, we need to pay attention to quality of bee products and specific protocole for each patient´s needs. To achieve the former, in the practical and private work of CENTROVETERA, we included the sourcing of the "best" beehive products and the handmade, artisan production of the "api-phyto-medical formulations" for animals, to walk from the pathology to health.
The presentation graphically shows examples of Integrative Treatment, including beehive products in different animal patients and applied by different ways. To simplify the presentation, the patients were ordered by Medical specialities, like Dermathology and Tegument Surgery (big wounds, skin and general dermatological disseases); Neurological / Neuromuscular diseases, Degenerative diseasses, Autoinmune diseasses, Reproduction, etc…

ARC (Apiceutical Research Centre): exploring a new generation of medicines from the beehive

J. Fearnley
ARC (Apiceutical Research Centre), Whitby, UNITED KINGDOM

Modern pharmaceutical medicine is in crisis. The search for the magic bullet is proving ever more elusive and inappropriate. Whilst the economically driven pharmaceutical juggernaut trundles on, the definition and meaning of medicine is being redefined – from targeted, synthetic and single molecule to natural, synergistic and holistic.
Science is turning back again to natural whole products with a new will to understand their complexity, intricacy and potency as medicines for man. We must thank the East Europeans for keeping alive a scientific understanding of bee medicines (Apiceuticals). Over 40 years ago the Kazan Veterinary Institute published research which showed that combining propolis with antibiotics increased the effectiveness of the antibiotics by up to 100 times. We had to wait till the 1990's before the West began to publish scientific papers.
Research into propolis has grown exponentially over the last ten years with Brazil, China, Turkey and India now major contributors.
Research in UK has grown steadily over the last 15 years particularly at the University of Strathclyde - Scotland. They have developed our understanding of how the chemical compounds collected by the honey bee from plants and trees within their local environment are transformed by the honey bee into a product able to provide immune defence for the whole superorganism.
The relationship between climate and the antibacterial properties of propolis is now more clearly understood. The discovery of anti-trypanosome chemicals in propolis in areas where there is sleeping sickness has opened up new research into Geographic Medicine.
Since 2011 ARC has been linking research activity round the world through a series of international conferences and more recently through the formation of IPRG (International Propolis Research Group)
ARC's Global BeePharma project further explores the concept of Geographic Medicine i.e. the chemical and biological activity of local bee products to local disease patterns in humans.
ARC has plans to build The BeeArc, a physical research centre and exhibition centre based in North Yorkshire UK and focussed on Apiceuticals – medicines from the beehive and Sustainable Beekeeping.
www.beearc.com

Therapeutic inhalation of beehive´s air - Characterizing the volatile components present in the air of beehive of *Apis mellifera* species

T. Guardia de Souza e Silva [1], O.V. Bustillos [2], P.O. Amaral [2], L.B. Motta [1], L.A. Praxedes [1]
[1] *Institute of Biological Sciences - Paulista University, São Paulo, BRAZIL,* [2] *Chemistry and Environment Center - Institute of Energy and Nuclear Research, São Paulo, BRAZIL*

The air inhalation from the hive of *Apis mellifera* honeybees species has been used therapeutically for several years. People from many European countries are currently being treated with hive air in naturalistic clinics. However, although in clinical practice this technique has positive results, the scientific knowledge about the mechanisms of its therapeutic properties are scarce, being necessary more research for characterizing the chemical composition of the volatile components present in the hive´s air and future identification of its pharmacological actions. This work has the objective of characterizing the volatile components present in the hive´s air of *Apis mellifera* honeybees species by the use of gas chromatography and mass spectrometry techniques. The hive´s air characterization is presented by the ratio of ions fragmentation (mass spectrum) of the founded compounds. Although future studies for specific molecular identification of each one of the characterized compounds by this work are still necessary, with our results it is possible to relate the chemical classes of the identified components with the chemical classes of the components founded in the bee products, including hydrocarbons, fatty acids, esters, alcohols, nitrogen compounds, aldehydes, terpenes, carbohydrates and phenolic compounds. Based on the comparison of the different chemical classes observed, it can be inferred that the air composition of the hive is formed by a collection of volatile components from each of the bees' products found in the hive. As a continuation for future works, the specific molecular identification of each one of the compounds characterized in this study could be carried out by comparing the mass spectra of the characterized compounds with the mass spectra of the possible molecules identified by the gas chromatography and mass spectrometry systems, followed by new comparative analyzes between the samples from hive´s air and samples of the specific molecules. After specific identification of each one of the volatile compounds founded in hive´s air, further studies on the pharmacological effects of therapeutic bee hive´s air inhalation may be carried out in addition to clinical studies in order to scientifically validate the practice of inhaling air from the hive for therapeutic purposes.

SYMPOSIUM 15:30-17:30
INNOVATIONS IN APITHERAPY
ROOM 517D

[Lead-off] Innovations in Apitherapy, a Review

S. Stangaciu
Romanian Apitherapy Society, Mereni Contesti (DB), ROMANIA

Due to the huge variability of the geo-botanical origin, characteristics, properties and uses of the beehive products, to the highly variable professional background and skills of the beekeepers (from unemployed to… academicians), to the huge importance of the beehive products for the human and animal health, to the globalization and Internet (with extremely fast communication), the Apitherapy offers a very important field for human creativity.
The huge number of diseases (over 20,000 according to the World Health Organization) and the fact that there are practically no 2 identical people, have forced the medicine oriented beekeeping and Apitherapy to adapt to the hugely complex individual needs of the human body structure and functioning, to the modern needs of Integrative Medicine, by creating hundreds of different apitherapy related preparations/ products and by adapting the treatment protocols to the individual needs of the patients.
If we analyze the preparation and products presented to the last Apimondia Congresses and Apimedica Symposiums we will see that they cover nowadays the needs of each part of the human/animal body, from eye drops with honey/propolis/royal jelly, nasal sprays, intravenous administration of honey and inhalations of beehive air.
In our presentation we will show the most impressive innovations in the field of medicine-oriented beekeeping and Apitherapy, thus stimulating the desire of the Apimondia participants to continue to create new preparations, new devices and new treatment protocols having the main goal to protect and/or restore the human/animal health, vitality and beauty.

[Lead-off] A review of 2018-2019

A. Thibault
Canadian Apitherapy Association, Toronto, CANADA

The Canadian Apitherapy Association is a young association that has been founded in 2016. There are 50 active members, individuals as well as organizations
Our mission is To understand, study, develop and organise all ideas, concepts, projects joining bees and health. By bees we understand, the bees, their role in nature and the bee products, by health we understand the definition of Dr. Bill Hettler from the National Wellness Institute: seven dimensions of wellness: physical, emotional, intellectual, occupational, social, environmental and spiritual. This definition

of Health is the reference in Canadian Universities since many years.
Our focus right now is networking, and education. Our team travelled in different countries to assist many events in France, Romania, Germany, Italy, Bulgaria, and Turkey. We met wonderful professors and speakers. With them, we already organized free monthly webinars for every one to continue to educate and promote apitherapy.
The goal of this lecture is to share projects, ideas, knowledges that inspired us.

Green Propolis (EPP-AF®) 80% powder extract, an innovative presentation with higher bioavailability effect

A. Berretta [1], R. de Souza [1], N. Ferreira [1], A. Buszinski [1], M. Araújo [1], J. Hori [2], T. Ramos [1], M. Tadini [1], F. Marquele-Oliveira [1]
[1] *Apis Flora Indl. Coml. Ltda., Ribeirão Preto, BRAZIL,* [2] *Faculty of Medicine from Ribeirão Preto, University of São Paulo, Ribeirão Preto, BRAZIL*

Propolis is a complex resinous mixture produced by bees. Chemical composition and biological action is variable considering type of bee, seasonality and vegetation available for bees. Because of this, it is important to standardize the product considering some biomarkers, in order to have a minimum reproducibility batch-to-batch, although, it is well known that, it is impossible to have identical batches of natural products. In this sense, EPP-AF standardization was previous obtained by Berretta et al. (2012), using majority green propolis as raw material. Propolis is also a very poorly soluble substance, and because of this, its bioavailability can be compromised. In order to try to solve this, our team develop two concentrated partially hydro soluble propolis powder (PPPM and PPPT), and these presentations were compared to others [alcoholic (GPFE) and aqueous extract (WSFE) liquid extracts, microparticles (MGPE) and lyophilized (LPE) propolis powders] against effectiveness and safety applying the methodologies: (i) in vitro antioxidant activity by H-donor activity using DPPH and chemiluminescence assays; (ii) cytotoxicity in macrophages and fibroblasts cell lines; (iii) anti-inflammatory activity by TNF-alpha and IL-6 cytokines; (iv) antibacterial activity against gram positive and negative strains. The extracts showed some interesting differences on their bioactive properties. DPPH scavenging activity results were 6 - 12 µgmL-1, with order of PPPM and PPPT>WSFE. In chemioluminescence procedure, IC50 values were 0.19 - 0.64 µgmL-1, and showed the activity order of LPE, PPPM, PPPT >> GPFE. In cytotoxicity assays, results demonstrated that for all samples, statistically difference when compared with control was obtained with concentration 125 µgmL-1 (except LPE, 60 µgmL-1) and for macrophage cell, all results were 250 µgmL-1. Thus, the extracts could be considered as safe. Considering anti-inflammatory activity, all propolis extracts were efficient in the reduction of IL-6. Antibacterial results demonstrated that the WSFE was the most effective one, followed by PPPM and PPPT in gram positive and negative strains. Studies of permeation were done with the measurement of Artepillin C, in order to compare PPPM with MGPE. The results demonstrated higher permeation of PPPM, suggesting that the innovative hydro soluble extract is able to demonstrate higher potential bioavailability.

Honey as the cure for the diseases of the twenty first century

B. Pradhan [1], V. Jessamy [2]
[1] *Indian Institute of Technology Bhubaneswar, Bhubaneswar, INDIA,* [2] *University of East Anglia, Norfolk, UNITED KINGDOM*

Honey has been used from the age of immemorial in traditional medicine. It was used to treat wounds, ulcers, throat, sunburn, eyes, and gut, etc. Thus, honey is gradually becoming part of modern medication derived from folk-medicine. In the traditional sense, honey was used for wound dressing, but with the advent of antibiotics, this was decreased. With the epidemic of antibiotic resistance bacteria, again honey has an excellent potency to be our savior in this hour of need. In addition, honey has antioxidant activity and anti-inflammatory action. Cancer, coronary, and neurological degeneration and most chronic diseases are the consequences of oxidative stress. Here we have tested and verified the bactericidal and bacteriostatic activity of twelve honey samples against different drug-resistant strains of bacteria, such as Klebsiella pneumoneae, Staphylococcus aureus, Escherichia coli, Salmonella typhimurium, Pseudomonas aeruginosa, Vibrio alginolyticus, Vibrio parahaemolyticus. The honey samples came from a conservation farm where nectar producing medicinal plants were selectively planted over a period of ten years. All the honey samples were able to inhibit both Gram-positive and Gram-negative bacteria at the concentration range of 10-15%. However, four of the samples stand out with respect to bacterial inhibitory ability compared to other samples. We have also found that three of our honey samples have higher bactericidal property compared to Manuka honey with bioactivity grade of 20+. Free radical scavenging ability of all the honey samples was studied by DPPH method. Interestingly, the honey samples having higher antibacterial property was found to have better antioxidant activity. We have estimated the biochemical composition of the honey samples through GC-MS and NMR. The honey samples with higher antioxidant and antibacterial activity are found to have diverse groups of phytochemicals present in them which are proven to be beneficial to human health Therefore, we propose that three of our honey samples have superior medicinal property to Manuka honey and have the potential to provide cures for the diseases of the 21st century.

Antimicrobial Activity of New Propolis Extracts: a Comparison

S. Keskin [1], M. Keskin [2], S. Kolayli [2], S.A. Karaoglu [3]
[1] *Bilecik Seyh Edebali University, Bilecik, TURKEY,* [2] *Karadeniz Technical University, Trabzon, TURKEY,* [3] *Recep Tayyip Erdogan University, Rize, TURKEY*

Propolis, a resinous mixture collected by honey bees, is not suitable for raw consumption. It should be extracted to convert it into consumable form. Many attempts have been carried out up to now to extract the compounds found in propolis. Most of these attempts

shared one thing in common that organic solvents like ethanol, ethyl acetate, hexane, chloroform, acetone etc. were used. In order to understand the chemistry of propolis, these solvents could be helpful but they are harmful for health. That is why most of them could not find application in the field except ethanol. The strategy of propolis extraction is mainly depends on the solvents used and ethanol is the first choice today and the market is dominated by the ethanol extract of propolis. Olive oil, water, fatty acids, glycerol as natural solvents and glycols, Gelucire 44/14 as synthetic solvents are also tested for preparing propolis extract. Although ethanol is reported to be the best solvent, it is also a limiting factor for applying propolis to certain areas like food and feed. Propolis is a natural antimicrobial agent and could be used as preservative in food formulations but its ethanol solubility, strong taste and aroma limits its usage in this area. There is an increasing need for new, healthier, compatible solvent applicable to food formulations. In this respect, we tested volatile oils for propolis extraction either alone or in combination. When used alone the solvent capacity of volatiles could not reached ethanol. A wide screening antimicrobial tests against some gram positive, gram negative and yeast like fungi were applied and it was found that antimicrobial activity of volatile oil propolis extracts was 10 times more than ethanol extract. In conclusion, volatile oil propolis extracts are good antimicrobial agents and they could be used as an alternative to synthetic drugs as a preservative or complementary medicine.

Effects of vegetation types and processing methods on physicochemical, phytochemical and anti-microbial activity of tropical honey samples invitro

C. Akachuku
Michael Okpara University of Agriculture Umudike, Umuahia, NIGERIA

The effects of the vegetation zones and processing methods on bactericidal and bacteriostatic properties of honey were investigated. The study aimed at comparing the physicochemical and phytochemical properties of honey samples obtained from the Rain forest, Mangrove swamp forest, Derived savanna, Montane forest and Sudan Savanna vegetation zones of Nigeria; determine the effects of methods of honey processing on the medicinal value of honey and evaluate the antimicrobial activities of the different honey samples. Honeycombs collected from hives in five vegetation zones were processed using three different methods namely: hand processing, floating and dripping. Factorial experimental design (3 x 5) was used for the study. Samples from the different vegetation zones were subjected to laboratory analysis. Data obtained were analyzed using ANOVA at 5% probability level of significance. Results obtained showed that honey sample from Sudan savanna had the highest specific gravity (1.4040 ± 0.01) and the lowest moisture content (14.59 ±0.003); Derived savanna honey had the highest protein content (0.47 %), followed by Sudan savanna honey (0.42 %) while the rain forest had the least amount (0.24 %) . On average, Mangrove honey had the highest amount of calcium (0.75 %), while montane honey had the least (0.03 %). Flavonoid content of honey samples from different vegetation zones and of different processing methods varied significantly at $P < 0.05$. The medicinal value was highest in honey sample processed by the floating method, followed by dripping and the least was that of hand pressing. Honey sample from montane vegetation was found to have bactericidal properties against Staphylococcus spp, Bacillus spp, and Escherichia coli, while honey samples from the same vegetation zones showed the highest phytochemical properties (0.96 % phenol, 0.16 % flavonoid, 0.46 % alkaloid), which confers the highest medicinal value on the honey. This was followed by honey samples from Derived savanna (0.86 % phenol, 0.14 % flavonoid and 0.42 % alkaloid) with the rain forest having the least values. Montane honey has the highest bactericidal and bacteriostatic properties, which makes it the best honey for pharmaceutical preparations.

Application of suppositories with bee propolis, royal jelly and homogenated drones larvae as efficient and safe apitherapy method in Ukraine

V. Knizhenko
Private Business, Brand Medok, Kiev, UKRAINE

In modern apitherapy, bee products are used in various forms: capsules, tablets, ointments, tinctures, and etc. One of the most effective and progressive forms for correct dosage is the application of suppositories. Suppositories (candles) can be rectal, vaginal and rectal-vaginal. Suppositories have several advantages: 1) Biologically active substances enter directly into general blood circulation (bypassing the stomach and liver) and can be absorbed at a high rate, which minimizes the risk of allergies and side effects; 2) The use of suppositories is simple, safe and painless; 3) The rectal-vaginal method of using suppositories combines the positive qualities of oral and injection methods of application of biologically active substances in a medical treatments; 4) In some cases (trauma, age, etc.), suppositories may become the only one possible method treatment.

Currently, these types of suppositories with various bee products are produced in Ukraine: 1) With bee propolis. Medical indications: prevention and treatment of gynecological, urological and proctologic diseases; 2) With Royal Jelly. Medical indications: immunity strengthening, restoration of work of urogenital, nervous and cardio-vascular systems; 3) With drones homogenate (homogenated drones larvae). Medical indications: restoration of the urogenital, immune and endocrine systems. Application of suppositories with such components as bee propolis, Royal Jelly or homogenated drones larvae now test in several clinics for true an efficient and safe apitherapy method. Several types of suppositories with bee products are unique, original, recently patented and produced only in Ukraine.

Also is important when in the suppositories include various phyto-components to enhance the therapeutic effect of treatment. This approach provides a high content of biologically active substances, as well as a significant synergistic effect from the mutual use of bee products and phyto-products for apitherapy.

It should be emphasized that suppositories with bee products are compatible with pharmaceutical treatment. Bee products are natural products with natural origin, therefore, contraindications to their use are minimal. At the present time the suppositories with bee products are widely applied in Ukraine. Practical application of suppositories in Ukraine is considered as an effective and safe method of apitherapy.

Allergomelissopalynology: a new process for the study of allergenic pollens of the honeys from around the world and its scope in practice for beekeeper and honey consumer

C. Nonotte-Varly [1,2]

[1] *Centre Hospitalier Marie José Treffot, Hyères, FRANCE,* [2] *Association Francophone D'Apithérapie, Lyon, FRANCE*

Globalization of the honey market involves knowing allergenic pollens (AP) of the honeys from around the world because to the best of our knowledge these are not well known and because allergic reactions are not uncommon among honey consumers.

226 honeys from 4 continents have been studied using a new process: allergomelissopalynology. This method describes the product typology and the geographical origin of the honeys, the spectrum of AP and the allergenic potency of the honeys using the allergenic potency index system (APIS). 95% of honeys contain AP, 7/10 in trace amouts, 3/10 with one or more AP > 3% with a maximum of 81%. In fact, AP in large amount are few. These are pollens of melliferous asteraceae (eg, sunflower, thistle, dandelion) and of non-melliferous plants (eg, asteraceae, oleaceae, gramineae). The allergenic potency of these AP depends on their mass within the honey and on cross reactivity between anemophilous and entomophilous pollens of tribes of, eg, asteraceae, gramineae, oleaceae.

This new method makes it easy to determine the AP spectra of the honeys from around the world and the allergenic potency of AP.

APIS could be used to inform the beekeepers appropriate combination of control measures to reduce, eliminate or prevent, as necessary, allergic hazards to allergenic pollens inclued in honeys, or to provide relevant informations to honey consumers to enable them to make better honeys choices, without risk to their health.

BEEKEEPING FOR RURAL DEVELOPMENT

09 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00
RESEARCH ON BEEKEEPING DEVELOPMENT ROOM 518

[Lead-off] Research on beekeeping development

N.J. Bradbear [1], J. Lowore [2]
[1] *Apimondia, Bees for Development, Monmouth, UNITED KINGDOM,* [2] *Bees for Development, Monmouth, UNITED KINGDOM*

Most of the world's poorest people live in rural areas: 80% of farmland in Asia and sub-Saharan Africa is managed by smallholders. In Africa alone there an estimated 41 million smallholders, and many of these people are beekeepers.
Recent years have seen increase in the numbers of beekeeping development projects that aim to help people move out of poverty by means of beekeeping.
What makes a successful project? What outcomes should be measured, and what evidence is needed to claim success?
There are many excellent examples of situations where beekeeping has enabled poor people to strengthen their livelihoods. Yet there are also situations where projects set off with unrealistic and exaggerated expectations, and when results are disappointing, do not review or publish what went wrong. Many beekeeping projects still focus on promoting inappropriate technology, to the benefit of the intermediary organisation rather than the intended beneficiary.
This introductory talk will consider approaches to ensure successful, long term, sustainable, beekeeping development.

Beekeeping, women and sustainable development

R.V. Regis De Sousa Gomes, T. Amaral De Barros, R. Silva Cabra, J.G. Souza Sales Albuquerque, H.L. Menezes Costa
University Federal Rural of Pernambuco, Department Animal Science, Recife, BRAZIL

The aim of this study was to evaluate the impacts of the establishment of a production apiary for the AgroFlores Peasant Women of Ximenes group (Mulheres Camponesas AgroFlores de Ximenes), in the city of Barreiros-Pernambuco-Brazil. The families of the AgroFlores Peasant Women of Ximenes group live in conditions of social and economic vulnerability, because the Zona da Mata of Pernambuco is a region that suffers from the secular cultivation of sugarcane, that has caused social, ecological and economic impacts. Initially interviews, seminars and workshops were presented to the peasants and the site planning for the apiary was discussed. For the acquisition of bees, bait boxes were distributed in the wild during the period of reproductive swarming, which made possible to acquire 15 colonies. The AgroFlores Peasant Women of Ximenes group is composed of 05 women, in which four are married and one is single. One of the women provided an area of one hectare near a reserve of Atlantic Forest for the establishment of the apiary. Sixty days after the establishment of the apiary the first honey harvest was made. It was observed that the bee management generated income and placed the women as active participants in their families' economy. It provided improvement in nutrition and health of the families for the consumption of honey. It promoted changes in the social role and the environmental perception of women, which encouraged them to plan the implementation of an agroforestry system to produce organic products integrated with beekeeping. And it boosted the development of environmental education activities in a multidisciplinary way, involving 29 children and young people of the community, collaborating environmentally with the conservation of bees. Although bee management is a heavy activity and the availability of bee supplies in the region is limited, women's teamwork and family involvement have provided positive impacts by reducing poverty, promoting food security, sustainable agriculture, health and well-being, as well as gender equality.

Approaches to targeting the poorest people through beekeeping in Amhara, Ethiopia

T. Gebey [1], J. Lowore [2]
[1] *Bees for Development Ethiopia, Bahir Dar, ETHIOPIA,* [2] *Bees for Development, Monmouth, UNITED KINGDOM*

Beekeeping can be undertaken by people who are economically poor because the input, labour and land demands are relatively low. This presentation concerns beekeeping in Amhara, Ethiopia, where despite these pro-poor factors, bees are more commonly owned by wealthier farmers. In a 2013 survey we learned that land-holding had a highly significant positive correlation with bee colony possession, indicating that wealthier farmers are more engaged in beekeeping. Government policy is to encourage beekeepers to change their beekeeping methods. One inevitable, but perhaps unintended consequence of this policy is that development projects therefore target existing beekeepers, and by extension the better-off, despite the government's poverty alleviation agenda. This was revealed during the 2013 survey, "Conventional agricultural extension didn't demonstrate its strength in ensuring emergence of new beekeepers rather building on pre-existing beekeepers". In this paper we explain how local-NGO Bees for Development Ethiopia implemented a project to increase household income through beekeeping 2015-2018. We targeted landless youth, people with few assets, those who worked for food and female headed households. The baseline mean annual income of the non-beekeepers was USD 300. We explain how the intervention was designed, the type of training provided and how the project nurtured a local beekeeping mentoring programme. The

project enabled poor households to keep bees and earn new income. Beneficiaries sold honey, using income for household expenses and children's schooling. Some were able to buy assets such as carts, solar equipment and ploughing oxen. By project end 469 people who had not kept bees before were selling honey (others had bees but had not yet harvested honey) and had increased their annual income by between 16% and 20% on baseline. Food security metrics showed a positive change. We learned that helping a non-beekeeper get started with bees can achieve great transformational change for a poor family. A beekeeping project for non-beekeepers requires a new approach, and it is harder to achieve change, compared to working with existing beekeepers. Training must be of high quality and continued for one year, with arrangements in place for new beekeepers to access help in their second year.

Fiji's Beekeeping Mentor Program

J. Caldeira
Fiji Beekeepers Association, Rakiraki, FIJI

Fiji is a small oceanic island nation without a long beekeeping tradition. In Fiji most beekeeping development projects have provisioned bee hives and equipment without sufficient training for long term success. The failure rate of new beekeepers assisted through development projects has been over 80%, as measured by the number of bee colonies surviving after the projects end. The two primary reasons for failure of beekeeping development projects in Fiji have been the focus on village ownership of hives instead of individual ownership and insufficient training in beekeeping skills.
To improve the success of beginning beekeepers, the Fiji Beekeepers Association, in partnership with the European Union-funded International Trade Centre, launched a Beekeeping Mentor Program where 20 beginning beekeepers were paired with local skilled beekeepers who were 'hired' to teach them practical beekeeping skills. The skilled mentors were incentivized with hive materials to teach beekeeping skills on an informal one-on-one basis over a one-year period.
The beekeeper-mentors understood beekeeping methods for the local conditions. The program provided the mentors with a list of skills to be taught in the field, including breeding bees, assembling equipment, managing bees for honey production, and identification of diseases. The first year of the program resulted in a high success rate for the trainee beekeepers, as measured by both colony numbers and a formal survey. Survey results also suggested several success factors for a mentor program, including proximity of mentor to the trainee, a preexisting relationship between mentor and trainee, and program monitoring. The results suggest that local mentors can substantially increase the success rate of new beekeepers and that inclusion of mentoring in a beekeeping development project is likely to improve overall project success.

How Can We Measure the Success of Beekeeping Projects? – A Case Study of a Cluster of Three Diverse Poverty Relief Projects and Their Local Lead NGO in Kasese, Western Uganda

R. Ridler, J. Ridler
Bees Abroad, Bristol, UNITED KINGDOM

Too many beekeeping projects are reported to have failed. Success is difficult to assess, and objective measurements can be simplistic. Success is best defined by the participants themselves. This paper presents a methodology for establishing success criteria of poverty relief beekeeping projects which could be used as a template for projects in other localities.
We describe a case study of a beekeeping course in Western Uganda attended by three diverse poverty relief projects, how they defined success and how measurements might be made.
The methodology used in this case study was a group interview, conducted in a discursive classroom environment and incorporated into a 2-day residential beekeeping course. There were three representatives from each of three beekeeping projects, initiated by the UK charity, Bees Abroad. One was a women's group, another a youth development group and the third a group of already established beekeepers benefiting from further development. A fourth group, with more of an overview, were the representatives from the delivery partner NGO, LIDEFO. The twelve participants represented a social, gender, age and educational cross section. All discussions were conducted both in English and the local language. The groups established their aims and objectives, differing in detail, but with many similarities. They then described ways that they, or others, could assess the achievement of each criterion, or if it was impractical or immeasurable.
From the study we suggest that a combination of quantitative and qualitative measurements that are defined by and can be measured by the participants themselves would best achieve the assessment of the success of a project. Examples of the former could be numbers/condition of hives or quantity of honey/products sold. One or more would be chosen and recorded on report sheets by the local leaders or beekeepers themselves, as appropriate. The interpretive measures of success, which really matter to people- whether children can be sent to school, or health improved, can only be achieved by questioning and observation of change in living conditions. A simple survey sheet could be produced, generated at the start of the specific project, for a suitable sample of participants.

Several key words used in beekeeping are racially charged: in order to make beekeeping and ecology more inclusive, we need to replace them

A. Roell, J. Van Wyk, M. Kirby, H. Whitehead
University of Massachusetts, Amherst, USA, [2] Washington State University, Spokane, USA

Several words used to describe honey bees directly reference the traumatic history of slavery in the United States. It is important to consider alternatives to these loaded and painful terms to make careers in the bee world more welcoming to people of color and to reach a broader audience as we communicate ideas about ecology and beekeeping. We borrow the concept of social reflection from the field of Environmental Sociology to explain how terms that humans use to describe nature often reflect the implicitly racist society in which they were developed (in the United States). This begins to explain how ecologists and beekeepers may use words that reinforce white supremacy without harboring racist intentions. Some of the problematic concepts and words include (1) Calling different types of bees "races" and using the word "interracial" to describe bee breed crosses. We investigate the biological and social connotations of the word "race" and consider alternative words that are more biologically accurate, like "sub-species" or "breed". (2) Calling hybrids of the sub-species Apis m. scutellate "Africanized". Scholars and the general public describe Africanized killer bees as "more violent" than European bees, and beekeepers worry that these Africanized bees will mate with their European queens. This description is dangerously similar to stereotypes against African Americans that white people have long used to justify racial oppression. We need to replace the terms "African" and "Africanized" in favor of more biologically-accurate words like "Equatorial". (3) Referring to bees that live inside the hive as "house" bees and bees that forage as "field" bees. The words "house" and "field" were used to distinguish the division of labor among enslaved people on U.S. plantations. Alternative words include "nurse" and "forager", which are already used in beekeeping. Recognizing how scientific discourse is constructed opens the space to imagine alternatives to critical terms that are rooted in the United States' history of slavery. These terms reinforce damaging racial stereotypes. Shifting our language, and selecting our words with care is a practical and powerful step we can take towards making bee science and beekeeping a more socially just practice.

Toward the protection of bees and pollination: the rise of global change science

A. Decourtye [1], C. Alaux [2], Y. Le Conte [2], M. Henry [2]
[1] *ITSAP-Institut de l'abeille, Avignon, FRANCE,* [2] *INRA, Avignon, FRANCE*

Over the past 30 years (1987-2016), bibliometric data shows a drastic change in the scientific investigation of threats to bee populations. Bee research efforts committed to studying bioagressors of honeybees (mainly Varroa sp.) were predominant, but now appear to be shifting from bioagressors to global change in the published literature. This rise of global change science reveals prevailing topics, for current and future years: climate change, landscape alteration, agricultural intensification and invasive species. We argue that with increased investment in applied research and development, the scientific, beekeeping and agricultural communities will be able to find management strategies for productive agrosystems and enhanced resilience of pollination and beekeeping. This implies the need for restoring and improving food resources and shelter of bees by ecological intensification of diversified farming systems, but also reconciling sustainable beekeeping with wild pollinator conservation.

BEEKEEPING FOR RURAL DEVELOPMENT

12 SEPTEMBER 2019

SYMPOSIUM
FOREST AND NATURAL BEEKEEPING

10:00-12:00
ROOM 517A

[Lead-off] Wilderness Beekeeping: Conserving a World Worth Living In

L. Sharashkin
HorizontalHive.com, Drury, Missouri, USA

Throughout most of human history, honey bees lived in the wild, and honey was a wilderness product. Today, beekeeping is seen as part of agriculture, yet agricultural landscapes become increasingly hostile to honey bees. Add to it the highly artificial methods of modern apiculture, and bee health issues do not come as a surprise, while pesticide-free honey is now a rare luxury.
But unlike farmers, most beekeepers do not own the land their bees forage on, and may feel powerless in the face of the ongoing environmental destruction. What can we do to reverse the trend?
Many beekeepers search for natural alternatives: they work with locally adapted strains; refuse to medicate their bees; raise them on a natural diet of honey and pollen; experiment with bee-friendly hive models and minimally intrusive management methods. But natural approaches cannot be successful in degraded habitat, so we must face the challenge of restoring health and beauty to our land. It has been said that “gardeners will save the world.” How about beekeepers -- are we up to this task?
When you practice natural beekeeping in a forest or other wilderness setting, you shield your bees from agrochemical pollution and give them the living environment they are best adapted to. You produce premium honeys from the richness of local plants free from pesticide residues. More importantly, bees help you create your livelihood while conserving the wild ecosystems they and us depend on. “Marginal” lands can often produce more economic value as bee pastures than through conversion to other uses such as animal grazing. And if we use a portion of our honey sales to purchase these lands and protect them, then beekeepers can accomplish much more than just producing honey and pollinating crops. We can help save the world full of blossoming landscapes, clear streams, buzzing bees, and children’s laughter. Can I count you in?

Forest beekeeping in Zambia’s miombo woodlands supports livelihoods and maintains forests

J. Lowore
Bees For Development, Monmouth, UNITED KINGDOM

Millions of very poor people live in Africa’s miombo zone, including nearly 200,000 who live in Mwinilunga and Ikelenge in Zambia. This study considers the local practice of forest beekeeping in Zambia, explores how selling honey meets the livelihood aspirations of beekeepers and investigates how they manage forest resources. A well-established honey buying company has been operating in this area for nearly 20 years. The study seeks to understand how the resulting increasing trade in honey is altering, if at all, the wellbeing of the primary honey producers and the way in which they value the forest. Field work in 2015, 2016 and 2018 comprised informal discussions with 19 groups of beekeepers, and a questionnaire survey involving 138 beekeepers, and 68 non-beekeepers in four sites. The study found that the practice of forest beekeeping is increasing, with income invested in education, in farming and as capital for other enterprises. Honey is often considered ‘the mother’ of other activities because no financial capital is required to generate this income. Self-reported measures of economic wellbeing showed beekeepers to be slightly better off than non-beekeepers. Although the forest is a common-property resource, beekeepers negotiate de facto rights to hive sites, and protect these sites from fire. This study suggests that unlike some NTFP harvesting, forest beekeeping is more than a safety-net activity. People are increasing their efforts, attracted by the rewards. Indications are that beekeepers have an economic incentive to take some forest management actions, and it is possible that these actions impact on a greater area of forest as the demand for honey continues to increase. The low productivity of miombo woodlands and soils do not offer a clear pathway out of poverty for the millions who live in and near the miombo zone. Yet the local practice of forest beekeeping, appears to offer some promise for forest-based poverty alleviation, whilst also avoiding forest loss. The approaches beekeepers adopt to protect their hives sites are locally driven responses.

Apiforestation in Appalachia: Reforestation on Surface Mine Sites

T.H. Potter
Kentucky Department of Agriculture, Frankfort, USA

Since 2008, I have worked with coal companies and a nonprofit, Green Forests Work, to reforest surface mine sites in Eastern KY with trees and forage that produce nectar and pollen. Over two million trees have been planted, and for ten years, I have coordinated a winter beekeepers school as well as coordinated pollinator education in the region. As the coal industry continues to decline in the region, many people are beginning to develop interest in beekeeping to provide a supplemental income. As the State Apiarist, I work with the Kentucky Department of Agriculture and the Governor’s Office of Agriculture Policy to find funds for schools and workshops. This presentation will

discuss the grants, the tree species, the projects, the honey marketing programs, and the setbacks of setting up a forest-based beekeeping industry in Eastern Kentucky. In addition, I have traveled to Africa, Australia, Europe, and South America to look at reforestation issues, and will incorporate those experiences into my presentation.

Apiculture and Traditional Forest Beeking in the Republic of Bashkortostan, Russia

R. Kuskildin [1], A. Murzabaev [2]
[1] *Bashkir State Agrarian University, Ufa, RUSSIA,* [2] *Russian State Agrarian University - Moscow Timiryazev Agricultural Academy, Moscow, RUSSIA*

The Republic of Bashkortostan lies at the intersection of Europe and Asia in the area of Ural Mountains where the pristine nature is preserved keeping the highest concentration of linden trees in Russia. Among the Russian population Bashkir people have been occupied by apiculture for the centuries. The harvesting of bees in the wild forest area has been developed and became the traditional for Bashkir people till nowadays. It is only in Bashkortostan where the population of Burzyan bee (Subtype of *Apis mellifera mellifera*) is protected by local law and the State Nature Biosphere Reserve "Shulgan-Tash" under the auspices of UNESCO have been created as well as the regional nature reserve "Altyn Solok", and the National Park "Bashkortostan".
This study is about the history of traditional beekeeping in Bashkortostan, its specifics. Secondly, we consider how the traditional beekeeping have been developed through the years focusing on its influence on the forest beekeeping in the republic of Bashkortostan nowadays. Thirdly, this study covers the preservation methods for the population of Burzyan bee, its importance as a result of negative effect of hybridization with subspecies, *Apis mellifera caucasica*, *Apis mellifera* carnica, *Apis mellifera* carpatica and others introduced from southern regions.

Bee farm

S. Poillion, D. Heijboer
Abeilles Libres, Lahitte, FRANCE

In the Hautes-Pyrenees, we are blessed with spaces that are more untouched than other parts of Europe; there are mountains and forests which provide plenty of nature for our bees. At the same time, much of the region is or has been used for extensive commercial agriculture, contaminating most often the areas used for human habitation. This leaves emptiness where there should be life. To counteract this, Ballot-Flurin Apiculteurs have developed a site called la Ferme des Abeilles Libres (the Independant Bee Farm), first in the world, that recreates the natural micro-biome previously lost to human usurpation. This differs from re-wilding, as the goal is to reconnect humans with the earth as well as the bees, rather than segregate us further. To overcome the inherent drive towards a protective separation, we have developed techniques for plant and land management that allow for communities based around symbiotic needs, rather than reinforcing the space between us.
Sabine Poillion will explain and present our site as it exists today, where we are advancing towards, and how we can recreate the same concept in different ecological areas.
Real pictures and results on honeybee health will be presented.

Development of organic beekeeping in the Gran Chaco, the strengthening of beekeeping organizations in the territory as a response to advances in transgenic echnology

R.H. Sayago, D.O. Garcia, H.C. Diaz, A. Sayago, I. De La Silva
Coopsol ltda, Santiago del Estero, ARGENTINA

The advance of the transgenic crops (soybean) and the extensive cattle raising pose to the Region of the great American Chaco a great problematic regarding its conservation and the sustainable development; the high rates of deforestation have an impact on the permanence of native peoples with large pockets of poverty and indigence. Cooperativa Coopsol, from the province of Santiago del Estero has generated a model of its own development, where the articulation with various actors, the boost to the growth of organic beekeeping and the same as traction of other productive activities, today allow organic beekeepers increase the income to remain in their territories, demonstrating that they allow the care of the forest against the advance of other production models, as in the case of Argentina, the model of extensive agriculture and the excessive use of agrochemicals to increase productivity.

Protected areas in Tanzania: Ideal for beekeeping, livelihoods support and home for pollinators

S.E. Msemo
Tanzania Forest Services Agency, Dar es Salaam, TANZANIA

Beekeeping plays a significant role in socioeconomic development and contributes directly to conservation of fauna and flora. Beekeeping is in Tanzania is forest based activity in most areas and few is practiced in farmlands and homestead. In the forests, beekeeping is mostly carried out in protected areas especially in forest reserves and game reserves of the mambo woodlands of central, western and southern highlands zones.

Management regime of protected areas falls under Tanzania Forest Services Agency (TFS) that is mandate to manage Forest Nature Reserves, Territorial Forest Reserves and Territorial Bee Reserves. Tanzania Wildlife Management Authority (TAWA) is mandated to manage Game Reserves and game controlled areas. Tanzania National Parks is managing national parks. Ngorongoro Conservation Area Authority (NCAA) is managing the Ngorongoro Conservation Area. Local Government Authorities in collaboration with communities are managing village forest reserves, village bee reserves and or wildlife management areas within their jurisdictions. Beekeeping activities are permitted in most of these protected areas, except in National Parks. The Authorities involved in the management of protected areas have been supporting local communities to engage in beekeeping

Beekeeping in protected areas act as an income generated activity that helps adjacent communities to improve livelihoods and minimize pressure on resources. Honey obtained in protected areas is multi-floral, natural and probably contained medicinal values and with a unique taste as well as free from pesticides and antibiotics.

Number of birds and insects (pollinators) are nesting as well as pollinate different flowering plants. Bee swarms have a tendency of migrating from one area to another (in the low lands to highlands and vice versa). These phenomena help beekeepers to site hives in adjacent swarming routes and obtain bee colonies. Swarming behaviour helps crop farms to obtain pollination services. Bee colonies also during dry seasons tend to abscond to the forest where they multiply and returning back to areas where beekeepers are siting hives.

Involvement of communities in the conservation activities and beekeeping is advantage in ensuring existing of pollinators, provision of nutrition (honey as food) and income generation through associate activities such as tourism, petty trade and employment.

CROSS-CUTTING SYMPOSIA

09 SEPTEMBER 2019

SYMPOSIUM 15:30-17:30

CITIZIEN SCIENCE AND BEES ROOM 518

[Lead-off] Best Management practices: a citizen science approach

D. vanEngelsdorp
Department of Entomology, University of Maryland, College Park, Maryland, USA

Citizen science is an efficient way to collect large amounts of data over a relatively short period of time. Here we explore this form of data gathering for the monitoring honey bee health. Surveillance data underpins epidemiological approaches to understanding organismal health, and in that regard the Bee Informed Partnership (BIP, beeinformed.org) has initiated several citizen science activities to help quantify bee health as well as identify factors that correlate to honey bee colony morbidity. Two of those initiatives: the US National Annual loss and management survey and the Sentinel Apiary program take very different approaches to surveillance. The first, an online and paper-based survey conducted over 11 years, has identified management practices linked with increased colony survivorship. While these practices differ slightly for beekeepers with different operational types (e.g. backyard beekeepers vrs. Commercial Beekeepers) some key management practices, like Varroa control, treatment of dead out colonies and equipment, and how new colonies are established were identified as practices, that when changed compared to “average beekeeper practice”, would have the greatest impact on colony survivorship. Subsequent field tests of these practices have validated these findings.
A second citizen science project is the sentinel apiary program, where participating beekeepers enrolled in the program monitor health in 4 or 8 colonies in a single apiary for a period of 6 months. Monthly summary reports allows the beekeeper to make timely management decisions. The data acquired from this system has huge epidemiological value as it permits us to retroactively compare colony health measures in populations of colonies that died compared to colonies that survived. This permits the identifications of which health measures. Landscape factors and management practices are most predicative of colony success. Hopefully, presenting results from this effort will help encourage others to either establish similar systems or participate in established systems.

The Sentinel Apiary Program: Collaborating with beekeepers to improve regional colony health and management

K. Kulhanek, D. vanEngelsdorp
University of Maryland, College Park, USA

The Sentinel Apiary Program is a colony health monitoring program run by the Bee Informed Partnership (BIP, beeinformed.org). Beekeepers enrolled in the program monitor 4 or 8 colonies in a single apiary for a period of 6 months. Colony monitoring efforts include monthly health inspections, monthly Varroa and Nosema samples, records of colony management and mortality, and optional hive scale monitoring. Varroa and Nosema samples are processed by the BIP diagnostics lab at University of Maryland, and a report with results is returned to the beekeeper within two weeks. This allows the beekeeper to make timely management decisions. Encouraging adoption of best beekeeping management practices is one of the primary goals of the Program. Participants say the Program holds them accountable for monitoring their colonies every month and thus improves their colony management and record keeping. Participants are also encouraged to share their reports with their beekeeping clubs and communities, empowering them to become ambassadors for good colony management.
Another goal of the Program is for Sentinel Apiaries to act as regional benchmarks for honey bee health. This is accomplished with the help of BIP’s online data portal, bip2.beeinformed.org. Here all Varroa and Nosema data are shared in an interactive online map. Pest loads are visible at state and county levels, allowing all beekeepers in an area to track how Varroa loads are changing around them. Users can also view the hive scale portal to see how weights change over time on an individual scale and state level.
Piloted in 2015, the Sentinel Apiary Program has grown from 21 apiaries to include 189 apiaries in 29 US states. Participating beekeepers have taken almost 7,000 samples and provided highly detailed longitudinal data. Data are currently being used to investigate trends in colony health in relation to factors such as management practices and apiary density. A tool developed in collaboration with NASA’s DEVELOP program will allow incorporation of NASA-EARTH satellite imagery into analyses. The Program will continue to grow in years to come and will enable better understanding and improvement of bee health and management across the US.

How clean is our city? A question for bee and human communities alike!

K. Smith [1], D. Weis [1], D. Hanano [1], J. Common [2]
[1] *University of British Columbia, Vancouver, CANADA,* [2] *Hives for Humanity, Vancouver, CANADA*

The most effective environmental assessments in cities not only require collaboration among scientists, engineers, and policy experts, but also the general community. Rapid urbanization, exploding human population, climate change, and the dynamic nature of cities in general, create critical scientific and societal challenges that highlight the need for ongoing and adaptive environmental monitoring in urban settings. Honey from *Apis mellifera* (Western honey bee) can serve as a biomonitor by elucidating small-scale pollutant distribution within a city. We have investigated lead (Pb) source apportionment in Metro Vancouver using honey, bee bread, and bees collected from urban apiaries in six local neighbourhoods. The results show that trace element concentrations and Pb isotopic compositions of these samples from various parts of Metro Vancouver differ systematically due to their proximity to anthropogenic metal sources related to land use, such as shipping ports and heavy traffic. Similar analyses on more traditional environmental proxies (topsoil and air particulates) have helped establish efficacy for our use of hive products as urban biomonitors. This study demonstrates the value of interdisciplinary collaboration when addressing issues related to environmental monitoring, urban ecology, and public health. Importantly, this work has benefited from partnerships with the general community and organizations including Hives for Humanity (non-profit, urban beekeepers with an extensive network of 'citizen' apiarists) and Metro Vancouver (official federation of twenty-one municipalities within Vancouver's metro region). This model provides compelling support for a citizen science approach where community members volunteer samples of their own backyard or rooftop apiary products to assess environmental conditions in addition to satisfying their own scientific curiosity. The results of this study provide a comprehensive baseline of trace element and Pb isotopic compositions in hive products, against which future results can be compared as population, land use, and regulatory policy change over the next century in Vancouver, BC.

Introducing the INSIGNIA project: Environmental monitoring of pesticides use through honey bees

N. Carreck [1], J. Amaral [2], C. Anagnostopoulos [3], H. Baveco [4], S. Bieszczad [5], D. Biron [6], R. Brodschneider [5], V. Brusbardis [7], L. Charistos [8], M.F. Coffey [9], C. Eulderink [10], A.R. Fernández-Alba [11], G. Formato [12], D.C. De Graaf [13], K. Gratzer [5], A. Gray [14], F. Hatjina [8], K.M. Kasiotis [3], O. Kilpinen [15], M. Murcia-Morales [11], M. Pietropaoli [12], M.A. Pinto [2], A. Quaresma [2], I. Roessink [4], J. Rufino [2], F. Vejsnæs [15], E. Zafeiraki [3], J. Van Der Steen [16]
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INSIGNIA aims to design and test an innovative, non-invasive, scientifically proven citizen science environmental monitoring protocol for the detection of pesticides via honey bees. It is a pilot project initiated and financed by the European Commission (PP-1-1-2018; EC SANTE). The study is being carried out by a consortium of specialists in honey bees, apiculture, chemistry, molecular biology, statistics, analytics, modelling, extension, social science and citizen science from twelve countries. Honey bee colonies are excellent bio-samplers of biological material such as nectar, pollen and plant pathogens, as well as non-biological material such as pesticides or airborne contamination. Honey bee colonies forage over a circle of about 1 km radius, increasing to several km if required depending on the availability and attractiveness of food. All material collected is concentrated in the hive, and the honey bee colony can provide four main matrices for environmental monitoring: bees, honey, pollen and wax. For pesticides, pollen and wax are the focal matrices. Pollen collected in pollen traps will be sampled every two weeks to record foraging conditions. During the season, most of pollen is consumed within days, so beebread can provide recent, random sampling results. On the other hand wax acts as a passive sampler, building up an archive of pesticides that have entered the hive. Alternative in-hive passive samplers will be tested to replicate wax as a "pesticide-sponge". Samples will be analysed for the presence of pesticides and the botanical origin of the pollen using an ITS2 DNA metabarcoding approach. Data on pollen and pesticides will be then be combined to obtain information on foraging conditions and pesticide use, together with evaluation of the CORINE database for land use and pesticide legislation to model the exposure risks to honey bees and wild bees. All monitoring steps from sampling through to analysis will be studied and tested in four countries in year 1, and the best practices will then be ring-tested in nine countries in year 2. Information about the course of the project and its results and publications will be available on the INSIGNIA website www.insignia-bee.eu.

Engaging bee-stakeholders for a bee-friendly Kyoto: A transdisciplinary research process

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The importance of honeybees and their direct and indirect services to human well-being earned lately increasing attention by media, policy-makers and researchers around the world. While a lot of that attention goes to the western *A. mellifera* and has a natural science angle to it, this presents a serious obstacle to improving the situation in the Japanese context of the world leading aging-society where a shift is happening: Professional *A. mellifera* beekeepers are retiring and declining, while retirees turn increasingly to *A. cerana* beekeeping as a hobby.
To grasp the challenges and potentials for beekeeping originating from the societal changes in Japan we chose a trans-disciplinary (TD) research process as method around the themes "A Future with Bees" and "Let's create a Bee-friendly City". This allowed not only beekeepers with diverse backgrounds, but also citizens and civil society groups to engage as bee-stakeholders. Furthermore the TD process shined a light onto the under-represented and under-studied humanities and social science aspects of honeybees and beekeeping while still integrating the natural science base, as well as focused on the lesser regarded eastern *A. cerana*.

The TD process started in fall 2017 and included in-depth interviews with experts and practitioners, participant observation, large-scale surveys of beekeepers (n= 386) and urban residents (n=700), as well as multiple stakeholder workshops. As TD aims at the integration of different kinds of knowledge and the cooperation between academics and non-academics in order to transform societal problems, this research process yielded 1) several research questions beyond the classical honeybee research with local, immediate policy relevance, 2) uncovered and documented previously ignored knowledge and practices of beekeeping, as well as 3) brought together actors beyond the typical 'beekeeping world' whose activities were already connected through the integrated reality of the ecosystem, but had socially not been in touch.

Adaptive Beekeeping in Canada's Far North through hive monitoring and a peer-to-peer knowledge sharing approach

E. Tardif
NA, Whitehorse, CANADA

My discussion will cover my approach in setting up trial polystyrene hives in two separate locations (5KM apart with a third location being added in 2019), using a data logging weather station, hive monitoring equipment (weight scale and temperature/humidity sensors), determining our local forage & bloom dates, using microscopy for Nosema testing, organizing a networking peer group of other beekeepers in the Yukon, Alaska and the Northwest Territories through social media and by conducting yearly beekeeping courses to existing and new beekeepers. The Yukon currently has no regulation regulating beekeeping (hive registration, disease monitoring, urban beekeeping) and it is therefore critical that all Yukon beekeepers are educated in hive management basics and disease identification and control. My goal has been to develop a better understanding of Yukon beekeeping methods (past ad present), identify best practices (to improve yields and winter survival of honeybees), identify our best forage for success (the best native and non-native pollen and nectar sources in the different parts of Yukon), and determine the best equipment to use for our cold climate. I now have 2 years of hourly weight scale data that has allowed me to determine our nectar flows (floral & honeydew) and typical bee foraging behaviours throughout the season. My internal hive temperature data allows me to better understand the impact of local climate on queen laying (start and finish) and to improve my overwintering approaches. It also provides internal clues on disease symptoms (e.g. fluctuating internal temperatures) which can be confirmed by actual hive observations. I now use hourly weather data (temperature, wind, rain) to graphically represent and compare my climate to other areas across Canada/World using typical bee foraging behaviours (FDH (>8C) Forage Degree Hours), NDH (>16C) Nectar Degree Hours, CDH (<8C) Cluster Degree Hours). For example the CDH value over time shows you the magnitude of "winter coldness", thus an indication of the energy requirements to maintain the hive over winter.
All this information has allowed me to develop and continue to improve my Northern Beekeeping Guide that I use to teach others to ensure the sustainability of Yukon beekeeping.

BeeKeep: a citizen science tool for bee monitoring and conservation

C. Barbiéri, S. Koffler, T.M. Francoy, A.M. Saraiva
University of São Paulo, São Paulo, BRAZIL

Due to the great importance of bees as pollinators, they are a priority conservation group. Effective strategies for the conservation of fauna and flora require a large amount of data on species abundance and distribution.Traditional methods of data collection on biodiversity require funding, time, and human resources aplenty. In recent years, citizen science approaches have become popular and generated impressive results in data collection and monitoring of biodiversity. Therefore, we have developed a citizen science platform (BeeKeep) which, through a mobile application allows citizen scientists to collect information on bees' distribution, nesting sites, and record interactions between bees and plants. Volunteer scientists photograph bees, bees' nests or interactions and send that information associated with GPS coordinates. The occurrence information is stored in a database compatible to the global citizen science platform iNaturalist, while the specialized data is stored in a personalized database. With these features, data on abundance, distribution and interactions can be collected by anyone with a mobile device with Internet access. Information of bees species composition in different landscape matrices will be analyzed from the collected data. The feature of species identification can also contribute to learning of bee identification by non-experts, since each register passes through multiple identifications and is validated by a specialist at the end of the process assuring data quality. This tool will comprise all bee species (native or invasive), with special functions for stingless bees, a diverse group of tropical bees with a growing beekeeping industry. Other features will be added in the application, including a tool for monitoring already-mapped bees, monitoring trap nests for stingless bees, and tools for stingless beekeepers to monitor the development of managed nests and their productivity. These new features will enable to extend the knowledge about the survival of natural nests, stingless bee phenology and obtaining data on the development of colonies. We believe this project has a great potential to bring the society closer to the knowledge-building process collaboratively, generating useful information both to increase knowledge about the biology of bees, but also to optimize the management of stingless bees.

CROSS-CUTTING SYMPOSIA

10 SEPTEMBER 2019

SYMPOSIUM 10:00-12:00

HONEY BEE NUTRITION ROOM 517A

[Lead-off] Nutritional health of honey bees in a changing world

A. Toth
Iowa State University, Ames, USA

Nutrition is fundamental to life. Without a balanced diet of adequate nutrients to support homeostasis, an organism's health and resilience to change can be compromised. The goal of my research program is to understand how interactions between nutritional stress and other environmental factors affect bee health and colony function. Working in the intensely agricultural Midwest, my research group studies how environmental factors such as floral resource dearth, pesticide exposure, and disease can alter honey bee nutrition, gene expression, and colony health. Field and lab experiments have shown that nutritional stress, such as feeding on low diversity pollen diets, can make bees more likely to succumb to other forms of stress such as viral disease. Interestingly, even when bees have sufficient food (i.e. are not suffering from a lack of calories), a lack of diet diversity is associated with large changes in global gene expression and increased disease susceptibility. Our studies on an apiary scale indicate that hives kept in extremely highly cultivated areas suffer from reduced nutritional health and queen fecundity at the critical pre-overwintering period, and that providing them with access to diverse natural habitat (prairie) can counteract this stress. These studies suggest that agricultural intensification, biodiversity loss, and global change are eroding bees' "nutritional resilience", thus making it more challenging for bees to deal with other forms of stress and contributing to colony losses and declines in bee health.

Brood rearing enhancing potential of manually packed pollen feeding in comparison with patty form of feeds

N.A. Mohammed, A.A. Al-Ghamdi, Y.T. Tena
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Dearth period pollen or pollen supplement feedings become very common practice to maintain and stimulate early buildup of honeybee colonies. Not only amount, type and consumption rate of feeds are important but the conversion and its nutrient availability are essential. In the current study we investigated the effect of feeding of colonies with pollen powder (packed on cells of drawn out combs) in comparison with pollen and pollen substitutes patty form of feeds on their dearth period performances. The study revealed that colonies given with pollen powder (packed on cells of drawn out combs) immediately converted the pollen into beebread similar to naturally collected pollen. The colonies in the packed-pollen group, reared significantly ($p<0.0001$) more brood and adult population than all other treatment groups. The colonies under packed pollen-powder group showed consistent increment and reared 86.3% more brood than the other groups, without having significant differences in their consumption rate. The possible reasons for better performances of colonies in packed-pollen group might be due to the conversion of the packed-pollen powder into beebread which allow better nutrient availability due to its fermentation by the nest microbes and further biosynthesis process. Moreover, the presence of beebread (from packed-pollen) adjacent to the brood combs represents the natural way of storing feed reserves which might have also contributed to stimulate brood rearing of colonies better than feeding in patty forms. Packed-pollen feeding could be better and alternative way feeding than patty forms for better maintenance and early population build of colonies.

The importance of nutrition for honey bee (*Apis mellifera*) health and colony performance during crop pollination

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When honey bees are being used for commercial crop pollination, food sources are often limited to a single crop. The lack of diversity in food sources can negatively impact bee health, resulting in malnutrition, decreased colony performance and ultimately poor crop pollination. One way to minimize such effects is to increase floral resource availability to bees during crop pollination. This can however be difficult to achieve under realistic field conditions and to be economically viable. Stored food sources collected prior to the start of the commercial pollination period could offer an alternative to maintain nutritional diversity. However, the effects of stored food on bee health remain understudied.

We conducted a field experiment using an agricultural setting to test the effect of food stores on bee health and performance. Forty colonies were divided into four different treatment groups that received nutritionally diverse food sources including mono – and multi-floral honey and pollen. The availability of pollen to bees was restricted during the pollination period by removing pollen from foragers with traps at the colony entrance. Parasite loads and foraging activity of workers and brood development were quantified. The type and amount of pollen collected by foragers, parasite load and brood development differed between treatments. Mono – and multi-floral honey and pollen impacted the metabolic profile of emerging bees. In summary we found strong support that stored food composition plays a crucial role on colony health, and our results offer possibilities to develop new management tools for sustainable bee keeping practices.

The effect of diet on the synthesis ability and gene expression of mandibular gland in honey bee workers (*Apis mellifera*)

Y. Chen, Y. Wu, S. Wang, H. Zheng, F. Hu
College of Animal Science, Zhejiang University, Hangzhou, CHINA

The mandibular glands of honey bee workers are key exocrine glands responsible for the biosynthesis of multiple royal jelly acids, which have important functions within the colony. However, our knowledge about the nutrition requirement of mandibular gland is quite limited. Here, we measured the effects of diet on 10-HDA and 10-HDAA synthesis ability, protein concentration and gene expression in mandibular gland. Laboratory reared worker bees were fed with pollen, soybean meal (protein supplement), stearic acid supplemented sugar candy, citric acid supplemented sugar candy or sugar candy only, and were sampled and analyzed at day 6, day 9 and day 12. We found that workers fed with pollen have the highest 10-HDA and 10-HDAA content on the head, and are significantly higher than workers fed with sugar only on day 6 and day 9, suggesting that imbalanced diet has a strong negative impact on the synthesis of fatty acids in mandibular gland. In the meanwhile, workers supplemented with protein or fatty acids have similar 10-HDA or 10-HDAA content with workers fed with sugar only, indicating that simply adding protein or fatty acids is not sufficient to meet the nutrition requirement of worker's mandibular gland. Interestingly, the expression levels of CYP6AS8, AOCX3, ACOX1 and CPT1 in the mandibular gland of nutrient deficit workers were higher than workers fed with pollen on day 6, then became similar on day 9 and day 12. Taken together, our results provide an insight for the nutrition requirement of honey bee mandibular gland and indicate that nutrient deficit might reduce the mandibular gland synthesis ability.

Role of micronutrients, gut microbiota and nurse bees for honeybee health

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Nutrition is a key factor for managed honeybee, *Apis mellifera*, health and macronutrients (sugar, fat, proteins) have comparatively been well studied. However, the roles of micronutrients (vitamins, minerals), gut microbiota and feeding of freshly emerged workers by nurse bees are less well understood. Here, we conducted a laboratory cage study over 22 days with freshly emerged workers (N= 1600) from three local colonies and investigated the effects of eight different treatments on worker longevity and body mass (N=40/group, day 7; with or without honey/pollen, protein-substitute lactalbumin, antibiotic tetracycline inactivation of gut microbiota, nurse bees, with eight cages/treatment with 25 workers/cage). The results show a significantly longer survival and a higher body mass in the workers supplied with honey/pollen compared to the groups without. In contrast, antibiotics treatment had a significant negative effect on both longevity and body mass, which may either be explained by inactivated gut microbiota or a direct effect of the antibiotics. Surprisingly, there was no significant positive effect of the presence of nurse bees on any of the parameters. The results clearly show the high relevance of full honey/pollen diet, thereby indicating the importance of a micronutrient-rich diet. The outcome further suggests beneficial effects of an intact gut microbiota on the workers. Moreover, the presence of nurse bees seems less relevant, suggesting that using bees hatched by themselves in incubators is an adequate approach for laboratory cage studies. In conclusion, improving nutrition via micronutrients/gut microbiota appears to be a promising way to promote honeybee health that merits more attention in light of multiple challenges.

Colony density and availability of resources limit honey yield

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[1] *Agricultural Institute of Slovenia, Ljubljana, SLOVENIA,* [2] *University of Ljubljana, Dept. of Forestry, Ljubljana, SLOVENIA*

Slovenian apiculture recognizes several single source honeys, none of them being of agricultural origin. Apiculture is woven into the fabric of Slovenian society which consequently boasts with high number of beekeepers and high colony density. An argument that environment carrying capacity has been (over)reached is often floated together with the question whether to limit the numbers of honey bee colonies in the vicinity of the nectar and honey dew sources. We have investigated impact of availability of natural resources and colony density on honey yield.

Data were collected over several years at key locations by means of "monitor" hives, equipped with scales. Locations were selected according to vegetation, ensuring the single-source nectar (or honeydew) flow. Source of the flow was recorded and verified by contract beekeeper. We investigated 1) relationship between abundance of the flow source expressed as quantity of wood stock within 3 km flying distance and net mass gain of the monitor colony during the flow; 2) relationship between colony count within 3 km flying distance and net mass gain of the monitor colonies.

Important single-source nectar flow in Slovenia is black locust, source of "Acacia" honey. We have determined the asymptotic exponential

relationship between its wood stock and mass gain: 20 x 10^3 m^3 of wood stock within 3 km flying range. Locations exceeding the limit did not improve mass gain of the monitor hive in normal years. Number of the colonies within 3 km radius did not influence the mass gain. Next, we calculated colony density in 3km radius as number of colonies against available wood stock in same radius and compared this new quantity against the mass gain. We confirmed exponential decay relationship: locations with density higher than 200 hives/10^3 m^3 of wood stock never exceeded 10 kg mass gain per colony regardless of the season. Locations with colony density less than 50 hives/10^3 m^3 wood stock gained more than 10 kg in 40 out of 44 cases, regardless of the year. We have determined similar relationships with linden/chestnut flow.
Our results show a limit on number of colonies at the location should be considered.

How to save the life of the prospective honey bee queen with royal jelly

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Honey bee royal jelly has long been considered only as larval food. However, as queen larvae are raised upside down in vertically oriented queen cells that open towards the bottom, the jelly also needs to adhere the royal larvae to the cell ceiling. This is where the acidic pH of royal jelly comes into play: only at a pH of 4.0 is royal jelly viscous enough to hold the larvae in their cells. We here show that royal jelly at pH 4.0 possesses a complex-tissue-like organization being similar to dense extracellular matrix of animals providing structural support. The main elements of the royal jelly “tissue” are proteinaceous fibril bundles that are largely destroyed upon increasing royal jelly pH to 7.0. The molecular basis of these fibrils are two proteins: major royal jelly protein 1 (MRJP1) and apisimin. Both proteins form at neutral pH a complex of four molecules MRJP1 and four molecules apisimin that polymerizes at acidic pH into long fibrous structures thereby increasing the viscosity of the solution.

SYMPOSIUM — 13:00-15:00
TREATMENT-FREE BEEKEEPING — ROOM 517A

[Lead-off] Breeding For Varroa Black Holes

J. Kefuss
Le Rucher D’Oc, Toulouse, FRANCE

During the 1980’s we helped develop chemical treatments for varroa mite control. Many of these treatments are still in use. Gradually it became apparent that mites would develop resistance to these chemicals. In the 1990’s we began screening and then selecting colonies for mite resistance (Varroa Black Holes). Eventually we developed three different selection methods: Bond, BAT (Bond Accelerated Test) and the Soft Bond Test. Regular advances in data collection and analytical technics are making these tests even more efficient to use.
The Bond test is “Live and let Die” survival. BAT involves placing highly varroa infested worker brood in breeder colonies to test their mite resistance. The Soft Bond Test selects first for production, then hygienic behavior and finally for mite resistance. Each test has its own specific advantages and disadvantages.
The goals of these tests are to maintain genetic diversity while conserving both short and long-term known and unknown mite resistance mechanisms. Colony mite survival is determined by natural selection. These are blind tests which means that you may not know why colonies survive. One advantage of blind testing is that unknown resistance mechanisms are able to express themselves. Once expressed they can be studied in detail and eventually used in breeding programs.
Economic comparisons between chemical treatments and breeding for mite resistance can be made since costs to produce breeder queens are easily calculated. This provides beekeepers with a valid decision-making tool to use in the fight against mites.

Towards Treatment Free Commercial Beekeeping in Greece

E. Koutouvela [1], N. Koutsianas [2]
[1] *Aristotle University of Thessaloniki, Department of Chemical Engineering, Thessaloniki, Greece, Thessaloniki, GREECE,* [2] *Symbeeosis, Athens, Greece, Athens, GREECE*

Greece has a big share in the European apiculture with approximately 1.3 million colonies. Greek rich flora and environment are potentially suitable for sustainable apiculture. Though apicultural conditions appear ideal, during the last few years there have been increased colony losses. During the last year, losses reached -and in many areas exceeded- 30%. Beekeepers and scientists accuse varroa and related viruses, other bee pests, nutrition, beekeeping manipulations and pesticides.
At the present study, we designed a scientific protocol suitable for the Greek apicultural conditions, aiming to ban the use of any chemical treatment and apply the optimum beekeeping manipulations, in order to limit colony losses and optimize the quality of bee products. We set up 3 experimental apiaries of 15 colonies each in three distinct locations of Greece (Samothraki, Andros and Chalkidiki), monitoring the colonies development, survival and levels of infestation for varroa, Nosemosis, American foul brood and other bee pests. The protocol

was based in scientific monitoring, regular sampling and analyses in order to evaluate the significance of each pathogen and determine the best method and time for its control. Colonies were headed by selected sister queens of *Apis mellifera* macedonica. Selection and rearing was conducted according to monitoring of colonies for their hygienic behaviour and their productivity. At the colony level we analysed the amount of the collected honey and pollen, the quality of honey and propolis, the strength of colonies etc. We tried to minimize any stress factors affecting the colony and we accommodated honeybees' homeostasis during wintering. After two years of application, no colony was lost and analysis of honeybees' products indicated honeys and propolis with superior characteristics (i.e. percentage of thyme pollen grains, higher antioxidant activity etc) compared to neighbour conventional apiaries used as controls. This study documented that the application of scientific beekeeping at the right place and time can result in maximum annual survival, and more qualitative bee product in Greece.

A side-by-side comparison of honey bee health in colonies managed using conventional, organic, and chemical-free systems

R. Underwood [1], B. Traver [2], M. Lopez-Uribe [1]
[1] *Department of Entomology, Penn State University, University Park, PA, USA,* [2] *Department of Biology, Penn State Schuylkill, Schuylkill Haven, PA, USA*

Honey bees are the most important managed pollinator for crop production, making the issue of colony losses a food security concern. Various practices are used to manage colonies, including conventional, organic, and chemical-free systems. An improved organic system is needed, as parasitic mite pressure and mite control chemicals are detrimental to honey bee health and sustainability. There is a critical need to develop scientific data that support organic beekeeping management practices with the goals of improving honey bee health and creating a profitable economic opportunity for beekeepers. This study is a stakeholder-driven, integrated systems-based project to rigorously test the effects of organic and chemical-free honey bee management systems on honey bee health by comparing the two systems to the control conventional management system. By measuring winter survival, honey production, varroa mite, nosema disease, and virus levels, and immunocompetence, we are able to determine the differences between the three systems. Our long-term goal is to generate evidence-based best management practices for a sustainable organic beekeeping system that will improve honey bee colony health, reduce environmental impacts, and increase economic returns to beekeepers.

Twenty Years of Commercial Beekeeping Without Treatments of Any Kind

K. Webster
Champlain Valley Bees And Queens, Middlebury, Vt, USA

Despite many contrary assertions, there are commercial beekeepers in North America who have successfully operated their apiaries without treatments of any kind for twenty years or more. Like all successful organic farms, each of these operations has developed its own unique management plan based on the local environment, and the resources available.

My own stationary apiary is located in a dairy farming region of the northeast U.S., producing honey, nucleus colonies and queen bees. The evolution of the apiary has been strongly influenced by an early connection with some of the early pioneers of modern organic farming in the U.S. and England. There were also three fortunate events that enabled the apiary to eliminate all treatments while maintaining productivity and profitability:

First was the inadvertent discovery that nucleus colonies could overwinter reliably outdoors, even through long, cold winters. This created a solid foundation for total self-sufficiency, production of surplus bees and queens, and serious bee breeding.

Second was the presence of tracheal mites for five years in my area before varroa mites arrived. The system of overwintering nucleus colonies already in place allowed for the rapid propagation of survivors without treatments, and resulted in an apiary with bees far superior to the original bees I had before the tracheal mite invasion. This and other lessons learned from tracheal mites were essential for our later success with varroa mites.

Third was the introduction of Primorski (Russian) bees into N. America. These bees had a more heritable tolerance for varroa mites than any other bees available here at that time.

Using breeding and management together, these three elements were combined into a profitable system producing bees, queens and honey without any treatments. Selection is based on a colony's total performance, and not on the monitoring of individual traits. Winter losses are higher than in the pre-mite days, but are usually easily recovered, and surplus bees have been sold for 18 of the last 20 years. This beekeeper now believes that the poisoning of the environment is the real existential threat to beekeeping--not varroa mites.

Healing Plants for Self-medicating Bees

F. Freeman
Centre for Urban Ecology, Humber Arboretum, Toronto, CANADA

Both native bees and honey bees have been experiencing serious declines over the past number of years. Faced with a variety of pest and pathogen challenges, and exacerbated by antibiotic resistance, the beekeeper looks to resolve these issues with new and improved medications and technologies. However, this approach overlooks, and can be at odds with, the innate immunity capacity and social immunity behaviours that bees have evolved including self-medication. This preliminary review of the literature looks at pharmacophoric behavior in bees, or the gathering and storing of plant materials to obtain beneficial phytochemicals and their consumption for medicinal purposes rather than for nutrition (pharmacophagy). Storage of these foraged products in the nest allows access to the "medicine cabinet" when a plant is no longer available. Plants contain secondary metabolites, or phytochemicals, which are defense mechanisms against insects and herbivores. Bees co-evolved with plants and can tolerate, and may benefit from, secondary metabolites which can protect these pollinators against pathogens and pests. There is increasing evidence that, when foraging, bees select for antimicrobial and other attributes. Plants also provide a niche for Lactic Acid Bacteria (LAB), some of which are closely related to those found in the gut of social bees. There is evidence for horizontal transmission of LABs to the hive environment and to colony mates through trophallaxis and other means. Although the mechanism is not well understood, LABs in the bee gut can induce an immune response in their host and different LAB strains may work synergistically to fight pathogens. A habitat rich in polyfloral forage increases the likelihood that not only are bees' nutritional requirements being met but also any medicinal needs. Some specific examples of medicinal plants will be considered.

Reintroduction of resilient honey bees into the wild in the Netherlands

T. Blacquiere [1], W. Boot [2], J. Calis [2]
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Very few (oral) reports on existing wild colonies in the Netherlands are noted, and a few examples known in recent years seem to be lost now. Honey bees in the wild have to cope with threats probably more severe than natural food scarcity periods and Varroa mites, being conservationists as well as beekeepers. In addition, not many suitable nest cavities are present due to management of hollow trees and increasing separation of conservation areas from the agricultural environment.
Since 2008 bees@wur and Inbuzz v.o.f. have allowed natural selection to shape two populations of honey bee colonies which survive Varroa without treating for Varroa infestations. The protocol used has been recently accepted for publication in Biological Invasions (Blacquière et al. 2019). Such populations might be an ideal basis for feralization and reintroduction of honey bees, provided enough nesting places are provided. Using colonies of our Darwinian selections to release swarms and by mounting 'Seeley' nest boxes in trees of a few conservation / nature areas, we aim to reintroduce vital populations of feral colonies in the Netherlands.

The Bzzness of Treatment-Free Queen Bees

S. Comfort
Anarchy Apiaries, Hudson, NY, USA

There are countless methods of raising queens and bees described in literature over the centuries, but very little discussion about how whole operations are organized and how the patterns of bee biology can influence the timing and methods to produce healthy colonies. We have a self-contained system of mating nuclei and cellraisers that yearly produces 3000+ queens, brood, bees, and honey with no treatments in simple, low-cost homemade hives. The simple, weekly schedule takes advantage of brood breaks during queen mating to suppress varroa mite levels and increase vitellogenin in the developing brood, which is banked for six days above a queen excluder to then be placed into queenless cellraising colonies. Boosting cellraisers and grafting on Mondays. Catching queens and banking brood from nucs on Tuesdays and Wednesdays. Putting ripe queen cells into the nucs on Thursdays. This biology-based, mechanical mite control in the nuclei can allow any operation the time and resources to start developing resistant stock. Anarchy Apiaries has been growing bees treatment-free since 2005 and continues countless experiments in making professional beekeeping simpler, easier, more affordable, and more fun again.

SYMPOSIUM — 15:30-17:30
HONEY MARKETING FOR SMALL-SCALE PRODUCERS — ROOM 518

[Lead-off] Creating a brand by telling your story: successful small-scale honey marketing

M. Denver
Apimondia, Bees for Development, Kingston, USA

Marketing your honey starts with creating a brand. In a small market, creating a brand can be as simple as telling your story. In this discussion, we'll share our story with you in hopes that it will inspire you and your honey marketing endeavors. We are a small beekeeping company located in the Hudson Valley of New York, USA. When we started our company it was our mission to help create a large sustainable beekeeping community in our area. Happily, after seven years, we've accomplished that and now provide continued support to established beekeepers and help new beekeepers get off to a good start. We are a one-stop-shop for beekeepers where we teach all aspects of beekeeping, sell all the gear needed, and the bees too. We have a strong honey market and sell almost all of the honey we produce at retail from our shop. We'll share our marketing and branding successes as well as a few failures we've had along the way in hope that you'll take away a kernel of our story that you can apply to your own brand creation.

Creating value for rural beekeepers

M. Muthu
T.i.A Bee Products Ltd, Quatre Bornes, MAURITIUS

The literature on beekeeping in Africa is sparse and usually refers to ad hoc projects in a few countries. Beekeeping is often discussed as a development tool to help smallholder farmers diversify their income.
A review of the honeybee and honey production in Ethiopia (Fikru, S. 2015) identified the inability to access markets as the most significant challenge faced by smallholder farmers. Remoteness, high transaction and transportation costs, lack of negotiating skills and bargaining power and poor market information are the underlying reasons standing in the way of market access. In addition, a project report published by the African Bee Resource Centre in 2017 (Carroll, T. et al. 2017), adds quality assurance, branding and traceability to this list.
To date no consolidated approach has been undertaken to: put African honey varietals on the map; highlight their unique identity and flavour; and highlight the communities behind their production. This is partly because the apiculture sector has historically been practiced using traditional methods in African countries and, as such, does not benefit from a well-informed and institutionalised framework in most African countries.
Our project set out to better understand the apiculture sector in African countries with a view to learn how to transform and commercialise beekeeping and make it an economically viable investment for smallholder farmers. A snapshot of the apiculture value chain in different country contexts showed that the sector is highly informal and contains several bottlenecks along the value chain. Most interventions to date have focused on the production phase of the value chain, with little money and energy left for the commercialisation aspects required to access premium market. Using this information, we embarked on developing a business model to commercialise beekeeping for African honey varietals.
Our business model contains components that are adaptable to local contexts while capitalizing on common aspects, such as suppliers' relationships, branding, and trade show attendance to put African honey varietals on the map. APIMONDIA 2019 will provide us with an ideal platform to showcase our work in Mauritius, Ethiopia and Madagascar, and the opportunity to network with potential partners from other African countries.

Granulated honey: From undesired product to winner of WBA Worlds Best Honey 2017

L. Fischer, Z. Knudsen
Lars and Zofuz' Beekeeping, Odense, DENMARK

In 2017, at the Apimondia Congress in Istanbul, we won the WBA honey contests and received the Carl and Virginia Webb Silver Bowl for the World's Best Honey.
The trophy winning honey was a creamy Danish spring honey predominantly based on nectar from rapeseed, hawthorn and stone fruits. It is the first time a granulated honey won this prestigious title. We will use our spring honey as case study for this talk.
In the process of preparing honey for the WBA contests, we have been studying and researching the chemistry and dynamics of crystallization in honey.
In this talk we will give an overview of what happens when a honey crystalizes and which tools we as beekeepers have at hand to influence and control this process. We will look at which factors induce and enhance crystallization and which factors inhibit and delay crystallization. Water content, temperature and the amount of crystal seeds in the honey are important parameters for the dynamics of crystal growth and thereby also for the quality of the final product.
Granulated honey is an exciting niche product for beekeepers in many parts of the world. It is a delicious food source which often stands out from the other types of honey at the shelf or at the market.
Producing the World's Best Honey has taught us a lot about treatment of granulated honey. We will share all our lessons and experiences with our fellow beekeepers.

Transformation of smallholder subsistent beekeeping through formal honey markets: evidence from Ethiopia

L.B. Shawul [1], W.A. Kebede [1], B. Taye [1], E. Mulatu [1], M. Belay [1], M. Kassie [2]
[1] *International Centre of Insect Physiology and ecology (ICIPE), Addis Ababa, ETHIOPIA,* [2] *International Centre of Insect Physiology and ecology (ICIPE), Nairobi, KENYA*

Ethiopia produces only about a tenth of its potential in crude honey and beeswax. A major reason for this wide gap is the subsistent mode of production with a few traditional hives around their backyards with very minimum management input. We present evidence from a pilot project that targeted delivery of a package of improved beekeeping practices can transform smallholder beekeeping from subsistence to commercial orientation. The package consists of selection of resourceful sites for setting up apiaries, enrolling of interested youth and women, organizing them into beekeepers' groups, delivery of beekeeping technical training, propagation of bee forages, delivery of beekeepers' starter kit, set up of honey and beeswax marketplace, promotion of market linkages and organic certification of the production and processing of honey. A pilot project at Tollay in southwestern Ethiopia involving an initial group of 120 youth and women beekeepers, organized into 10 primary cooperatives, led to a five-fold increase of their incomes from beekeeping compared to control households.

After six years of promotion the number of participating farmers increased to over 900, 72.4% of which had fully adopted the introduced frame hives whereas the rest 27.6% opted to continue to use traditional along with frame bee hives. Local and introduced bee forages got established to support expanding apiaries. Upon organic certification of the production and processing of honey, and a markup price of 20% was agreed for the packed table honey with the organic certification seal, the marketplace owned and managed by the cooperatives' union processed and packed 20 tons of labelled table honey. Also, crude honey harvested from frame hives fetched more than double the price of crude honey of about US$2.05 per kg. Subsequently at least a few hundred indirect beneficiary beekeeping farmers adopted aspects of the improved beekeeping practices and established proper apiaries. The success in this pilot project led to initiation of an up-scaling project in another region to support 10,000 needy youth and women, and proposition of another project to overlay managed beekeeping for pollination service and hive products in a large area producing organically certified coffee.

Madhuban: Honey Brand Shaping Honeybee Economy of Rural Maharashtra

S. Waslekar [1], V. Chordia [2]

[1] *Officer On Special Duty, Chairmans Office, Mumbai, INDIA,* [2] *Chairman, Maharashtra State Khadi And Village Industries Board, Mumbai, INDIA*

Maharashtra State Khadi and Village Industries Board (MSKVIB), is the provincial level authority solving three challenges in its effort to co-create a sustainable rural honeybee economy:
-Creating market channels for honey producers
-Institutionalizing honeybee products for benefit of rural industry
-Providing consumers organic honey sourced from within the State of Maharashtra
These challenges address concerns of environment, economy and society pertaining to a structured honeybee economy. Small producers of honey or honey entrepreneurs are found across Maharashtra. MSKVIB registers their details, purchases their honey, conducts quality tests and sells the honey across Maharashtra and India under the brand name, Madhuban. Hereby, through means of working directly with honeybee producers, scientific community, rural entrepreneurs and other related government departments, Madhuban has ensured sustained employment and dignity of rural honey entrepreneurs.
Once, honey producers, working with MSKVIB clear introductory formalities, then, they are provided training on scientific means of honey collection, awareness on how their honey will be marketed and follow up on future cooperation. In accordance with principles of rural industry development, MSKVIB has created an ecosystem for rural industry products.
Madhuban is sold in Mahakhadi retail store. Mahakhadi is MSKVIB's retail outlet that sells Madhuban brand, other Maharashtra honeybee economy products and wide range of rural products originating from Maharashtra's rural industries. This is exactly where the story of the rural honey producer is brought to the surface and their efforts are recognized by urban population. MSKVIB, has most recently given master franchise rights of Mahakhadi, enabling the store to become a chain and multiply across Maharashtra, India and world over.
The implication of such business strategy is multi-fold. First, it provides the honey producer a sense of greater community belonging. Second, it ensures sustainable business practices through demand for Madhuban in Mahakhadi stores in India and abroad. Finally, and most importantly, it weaves the story of rural Maharashtra for the benefit of beekeeping for rural development.

Application of Flow® technology to optimise returns for small honey producers

E. Grace, S. Anderson

Flow TM, Bangalow, AUSTRALIA

The 'local food' movement has grown substantially in the last decade with consumers willing to pay a higher price for quality local produce. More consumers are seeking to connect with their local farmers, and experience true paddock-to-plate produce. The Flow Hive presents a valuable opportunity for small producers to optimise their brand distinction and supply high-quality, high-value boutique honey. Flow Hive honey stands out as an unprocessed, raw product presenting opportunities for new marketing approaches.
Reports from consumers and apiarists that Flow Hive honey tastes better, "..the superior flavor quality of the honey has won many a loyal customer at my local farmer's market." Samuel, Wisconsin, USA, are supported by a recent study finding Flow Hive harvested honey retained more fresh, floral attributes than equivalent conventionally harvested honeys[1].
The ease of harvest enables beekeepers to demonstrate direct harvesting at markets or community events. While in-field harvesting is enabling larger operations to more readily isolate individual high-value honey crops. Case studies will be presented of innovative small- to medium-sized honey businesses using Flow Hives in various settings to maximise returns on their honey.
Sensory descriptive profiling of Flow® honey compared to honey extracted using conventional methods. S. Olarte et al. 2017.

Social Media Networks Strengthen Small-scale Honey Producers in Australia

B. Moore

Ben Moore, Melbourne, AUSTRALIA

Australia's geography is vastly remote and unpopulated, with 71% of the population living in capital cities (ABS 2016). And while the rural economy is experiencing a boom due to growing exports and free trade agreements (Quinlivin 2017), small farmers are not always included

in this prosperity. Small-scale honey producers can be represented in this group, with alternate methods required to communicate and market their product. Research has shown that there is an increase in the use of social media platforms within agriculture worldwide: however, there are still challenges for this cohort, including illiteracy, limited participation and lack of quality control (Barau and Afrad 2017). With the right approach, these avenues can provide access to sales, a platform for business promotion, and inclusion in the broader conversation around beekeeping; this is especially useful in Australia, where the vastness of the country, and subsequent differences in environment, produce specific honey products with unique characteristics. And while previous "word-of-mouth" marketing methods could prove difficult in rural and remote areas, online media such as Facebook, Instagram, Twitter, blogs and forums have enabled small-scale bee-related business to flourish despite considerable geographical handicaps. This may be in the form of online sales, or promoting distribution points for their product, such as local retailers, "honesty-system" stalls on properties, and farmer's markets. Furthermore, a positive, environmental and political use of social media networks has also proved beneficial for local honey makers—they have been able to expand their sales and profiles by educating the population about bees and their connection to biodiversity. This was proven in a recent media debate, that raised widespread community awareness of the potentially unethical practices of Australia's largest honey producer. Small-scale producers received attention for their contribution to this conversation, especially via their social media channels, increasing their sales and professional profiles (Elmas 2018). In turn, small-scale honey producers can utilise high-quality social media marketing strategies to access an already prevalent "buy local" ethos in Australia, promoting their unique product, and increasing their chance of success.

CROSS-CUTTING SYMPOSIA

12 SEPTEMBER 2019

SYMPOSIUM 13:00-15:00

BEEKEEPING WITH LOCAL & INDIGENOUS BEES ROOM 517A

[Lead-off] Beekeeping with Local and Indigenous Bees - An Overview

G. Otis
University of Guelph, Guelph, CANADA

Beekeeping done appropriately has the potential to enhance incomes and well-being of rural people. Depending on the local species/race of bee, floral resources, regional farming systems, and societal norms, that potential ranges from slight to great. Correspondingly, the potential of success for projects designed to improve honey production also ranges from slight to great. Surprisingly, beekeeping development projects may do more harm than good if the people spearheading them fail to consider the full range of factors impinging on the project. The success rate of beekeeping development projects is poor; failure to work with locally adapted bees is one factor that contributes to their poor success. In Asia, the indigenous hive bee, *Apis cerana*, is common. It can generally be kept with few inputs and low financial risk to the beekeeper. Hives can be made from scraps of wood, clay, even mud. If mistakes are made and the bees die or abscond (abandon their hive), more can be readily obtained for free from the surrounding environment. Some races of bees produce smaller crops of honey and are more difficult to manage than others, but financial profit is possible from all due to low start-up costs.
Because European honey bees are exceptionally well understood, beekeeping "experts" from Europe and North America who generally have little experience with *Apis cerana* often promote European *Apis mellifera* in development projects in Asia. Imported packages of bees are extremely expensive. The imported bees require extensive feeding during resource poor periods and costly treatments to control Varroa and Tropilaelaps mites. If a colony is killed by mites or becomes queenless, the cost to replace it (or to pay off the loan for its purchase) may be more than the annual income of the beekeeper! The financial risk to the rural farmer is huge. Even though potentially high yields of honey may be possible, high costs coupled with high risk of colony loss makes small-scale beekeeping with *Apis mellifera* impractical. The presentation will also touch on beekeeping with honey bees in South America and with stingless bees throughout the tropics.

Shifting Goal post: From Promoting Frame-hives as Magic bullet to increase apiculture production to Importing Queen and Bee-breeding in Uganda

B.D. Biryomumaisho
The Uganda National Apiculture Development Organisation, Kampala, UGANDA

While growing, I kept bee in traditional hives- found the culture. While at University -2004 taking beekeeping course-unit, I discovered other hives Kenya-Top-Bar and Langstroth. I got convinced that Langstroth followed by KTB are a-must-do following the theory acquired in class and internet publications. I thought of forming national beekeepers' association to cause change, fortunately TUNADO-national member beekeepers body existed it was dream come true. To join I mobilized community formed FEDN and secured membership. In 2008, was voted as TUNADO-board member. I secured beekeeping project for FEDN establishing demonstration centre with langstroth and KTB from Golden Bees procured their services to train beekeepers. Under took one month-internship at NAC -was using all the technologies, only local hives and KTB, were performing. I applied acquired skill but was successful with KTB only. Questions started running in my mind why do equipment traders and professional beekeepers from developing countries promote langstroth and not traditional or KTB?. Luckily, enough there were a number of project by different international organisations majority promoted modern-beekeeping (hive-technology) to cause change while a few promoted traditional hives. We received expert from developed countries trained in modern-beekeeping. In 2008 an MSP involving beekeepers, Government, private sector and development partners was formed since then hive technology dominates MSP agenda. Organizations emerged to producers and sell langstroth. Different projects bought and distributed langstroth to farmers. Whenever farmers are tasked on productivity of three technologies no answer for langstroth. Promoters of langstroth blames farmers of limited skills, hobbyist, and laziness while beekeepers say the technology does-not work. Langstroth promoter are challenged to put up demonstration under both their control and that of farmer. They never go that way but when in trade shows, workshops, they promote langstroth. The most recent development in Uganda are langstroth promoters changing goal post to blame it on indigenous honeybees and demanding immediate importation of queen bees and using the imported bees to breed as a means to boost production. The questions is it bee, beekeeper, technology or promoters with problem.

Defining the "local" bee: between globalization and regional adaptation. An example from French apiculture

A. Fortier [1], L. Dupre [2]
[1] *INRA/sae2/sadapt, Ivry/sur/seine, FRANCE,* [2] *INRA/sae2/cesaer, Dijon, FRANCE*

Thanks to its small size, among other factors, the honeybee is a creature that is easily transported and exchanged. A significant market in honeybees exists, one that dates back in France to the mid 19th century, with the importation of the Italian bee, celebrated for its productivity. In the absence of any formal breeding program adopted by the French apiculture sector, this market has continued to expand over the decades. Although originally based on an indigenous breed of honeybee adapted to local environmental conditions, French apiculture has not escaped the effects of market globalization, resulting in significant genetic mixing. The development of new honeybee strains, such as the Buckfast bee, has likewise contributed to widespread interbreeding. In the current context, with disease pressures increasing as a side effect of substantial honeybee imports and other, equally serious threats weighing on the sector—including climate change, the loss of floral diversity, the use of pesticides harmful to bees—the question of genetics is again on the agenda. Two trends are emerging in response to these threats and uncertainties. The first favors a return to the use of “local” or “regional” honeybees and other native bees; efforts are underway to protect the genetic resources of these heritage strains and species. The second seeks to explore other genetic combinations, with “local” bees contributing to the mix via natural, spontaneous crossings. A tension has emerged between those favoring the “black” bee—hardy, local, but considered by some to be aggressive—and those preferring the “yellow” bee (now called the Buckfast)—productive, gentle, but coming from a geographic, technical, and commercial “elsewhere.” We suggest that these two positions point to the same underlying question: how does one best align a type of bee, a local area, local resources, and a specific way of understanding and practicing apiculture? In this presentation, we will also emphasize the semantic slippages that characterize the use of these different categories: “local” bees, the “black” bee, the “native” bee, “Buckfast,” etc. Our analysis is based on a sociological study relying on interviews conducted in two French regions, the Parisian region and Burgundy.

Beekeeping - the Zimbabwe perspective

J. Gowe
Sweet Maungwe Honey, Rusape, ZIMBABWE

Zimbabwe has practised beekeeping since the 18th century; paintings in the Matopo Hills matebeland province of Zimbabwe depicts the San people practising beekeeping. Currently four species of bees are prevalent : *Apis mellifera* scuttellata, litoreia, adesonii and sting-less bees - *Trigona*. It is assumed the litoreia and adesonii found in the coastal regions might be breeding with the scutellata and producing hybrids .Honey production historically was for home consumption but with the commercialisation of the sector and healthy living to reduce life style diseases honey consumption is on the increase country wide. 80% of the beekeepers use traditional hives and 20% using a mixture of langthorth and Kenya top bar hives.
Beekeepers have embarked on keeping the sting-less bees in modern hives and experience high levels of absconding. Indigenous knowledge passed on from generation to generation has been useful in some areas such as various plants used by rubbing on ones body when harvesting instances where protective clothing is not available when harvesting from colonies of *Apis mellifera* Traditionally eating of bee larva mixed with combed honey for the men in communities has also been passed on from generation to generation.
However it has been noted that some of the traditional hives are prohibitive to farmers to conduct inspections hindering them from being able to control pests such as hive beetles or monitoring honey production which affects the yields. From training conducted by the author, a knowledge gap of market access, apiary management, grading, veld fires have been major challenges the beekeeping sector faces in Zimbabwe. However with the gradual adoption of modern methods of beekeeping a steady increase in production is noted in some areas. Efforts are being made by the national apiculture platform to promote more training, collective collaboration of sector players to improve production, by sharing of best practices, capacity building, lobbying policy makers so that Zimbabwe honey can be sold globally.

Honey Bee Impact: Using sustainable beekeeping for community-driven development

K. Wolf [1], G. Perilla [2]
[1] *Champlain College, Burlington, USA,* [2] *George Mason University, Fairfax, USA*

Honey Bee Impact is a non-profit organization that combines beekeeping research and training with global education in order to achieve community-identified sustainable development goals. With ongoing projects in Colombia, Panama, Puerto Rico, and Peru, our organization delivers bee-related programming in diverse settings and through customized approaches. This talk will focus on our research efforts in the Peruvian Amazon to manage endemic, stingless bees, including Melipona eburnea, M. elota, M. favosa, and M. compressipes, as a source of medicinal and marketable honey for rural rainforest communities. Since 2012, we have collaborated with and experimented alongside community beekeepers on sustainable beekeeping and honey harvest techniques that mostly encourage the transition from tree hollow hives to vertical hives. The vertical hive structure has numerous benefits, including a less invasive entry into the hive for honey harvest and the ability to make splits for further colony propagation. After identifying ideal tray depths and species-specific preferences for wood varieties over several years of experimentation, many colonies have been successfully established in the community and interest in keeping bees continues to grow. Community members report a vast array of medicinal uses for their honey (treating cataracts, respiratory illness, intestinal parasites, and infertility, to name a few) and can sell and trade the high-quality honey harvested from vertical hives for significantly increased value. The integration of this work with the global studies curricula at our home institutions adds to the value of this project and has served as a model for creative interdisciplinary work in the realm of international development, particularly by offering flexibility in the types and approaches of development work our students can engage in. Additional updates on colony success, hive design modifications, pest management, honey processing, and beekeeper recruitment will be reported from our Summer 2019 trip.

Traditional uses and relative cultural importance of stingless bees (Hymenoptera: Apidae, Meliponini) in Nepal

C. Bhatta [1,2]
[1] *University of Kansas, Lawrence, KS, USA,* [2] *Jefferson College of Health Sciences, Roanoke, VA, USA*

Indigenous and non-indigenous peoples in tropical and subtropical areas of the world use stingless bees for diverse purposes. There are no documented published records on traditional uses of stingless bees from Nepal, however, scattered information indicates that people keep stingless bees and utilize its hive products in different region of Nepal. We conducted ethnographic research across the Terai and Pahad regions (9 districts and 7 zones) of Nepal through field visits and informants interviews. Eighteen specific uses of stingless bee products in food, medicine, crafts, and religious beliefs were recorded from nine districts and seven zones of the country. People largely exploit these resources through a destructive extractive management practice. We document for the first time local indigenous nomenclature, uses, and its traditional management practices of only one provisionally identified species *Tetragonula iridipennis* (Smith) in Nepal. In addition, we determine the relative cultural importance of this species for each ethnic community (Chhetri, Brahmin, Tharu, and Kirat) that participated in the study. We also discuss the conservation status and future directions for the sustainable use of this bee species in the country.

Locally adapted bees perform well and appear varroa tolerant

P. Kryger [1], F. Zagni [2], N. Lanteri [2], C. Costa [3]
[1] *Aarhus University, Slagelse, DENMARK,* [2] *Apiliguria Italy, Airole, ITALY,* [3] *Apiliguria Italy, San Remo, ITALY,* [4] *Centre For Research Agriculture And Environment, Bologna, ITALY*

Italy is home to four subspecies of honey bees. Along the border to France a population of A. m. mellifera occurs. The genetic characteristics of population are described based on new data using single nucleotide polymorphisms (SNPs). The local honey bees appear relative pure, but hybridisation with the more common subspecies of Italy, A. m. ligustica is observed. However, the ApiLiguria beekeepers want to preserve their local heritage and are actively avoiding importing stock from other parts of Italy.

The local honey bee benefit from its adaptation to the climatic conditions in the region and the associated flowering patterns. The bee is able to survive equally well in the wild and beehives. The mountainous region does reduce gene flow with other populations helping to preserve the characteristic of the bees.

Observing how well the wild bees survive in rock caves, hollow trees and in buildings out of reach have inspired the local beekeepers to cease varroa control in their colonies. A large fraction of the ApiLiguria beekeepers indicate that they haven't treated their bees to control varroa for ten or more years. The healthy interaction between beekeeper management and locally adapted bees, seems to have let to the development of varroa tolerance in the region.

SYMPOSIUM 13:00-15:00
INNOVATIONS FOR MONITORING COLONIES ROOM 517D

[Lead-off] Got sensor, now what? Extracting information from continuous data

W. Meikle
Carl Hayden Bee Research Center, USDA-ARS, Tucson, AZ, USA

Just as continuously monitoring pulse, respiration, blood pressure and other variables is becoming commonplace for humans, continuously monitoring physical variables associated with honey bee colonies, including weight, temperature, respiratory gases, sound, and forager traffic, is becoming feasible for most researchers as cost and size of sensors decrease while their precision and capacity increase. This approach to honey bee colonies is logical because obtaining data from sensors reduces or eliminates colony disturbance, and provides new kinds of information about colony health and activity. The focus of our research is on extracting information from such sensors and relating that information to colony activity and growth. These points will be illustrated with data from recent field and laboratory experiments involving manipulative experiments exposing colonies to sublethal pesticide concentrations as well as monitoring commercial colonies exposed to different landscapes. We have found that sensor data can detect effects on colony collective behavior and health in situations that periodic hive inspections or standard analyses of individual bees do not.

RF Doppler Sensor for Assessing Beehive Health

H. Aumann, N. Emanetoglu
University of Maine, Orono, Maine, USA

In Maine, the pollination of commercial blueberries is carried out by honeybees from hundreds of hives. A low-cost activity sensor for unobtrusively monitoring the health and productivity of a beehive is presented. The sensor is portable and does not require any disassembly or modifications of a hive.

The sensor is based on a low-power 24 GHz Doppler radar module that is commonly used in automobile collision avoidance systems. With

some modifications it is capable of detecting honeybees in flight at a short distance. The output of the Doppler sensor is an electronic signal the 0 to 600 Hz frequency range.
Beehive health and productivity are assessed by monitoring the flying activity of bees arriving at and departing from the hive entrance and by comparing it with the activity in front of other hives. To reduce the large volume of data to a single meaningful index, the level of activity is quantified by the average root-mean-square (RMS) power in the Doppler spectrum. This index is collected every 2 minutes during daylight hours and transmitted to a central node by a wireless network.
Data from three instrumented beehives collected during the summers of 2017 and 2018 are presented. The activity indices were found to be highly correlated with environmental effects, such as temperature and solar radiation. They were also indicators alerting the beekeeper to immediate intervention, such as hive failure, absconding, swarming and robbing. The technique was validated by comparison with visual hive inspections.

Shotgun metagenomics and targeted analyses of honey derived environmental DNA define novel strategies to monitor honey bee pests, parasites and pathogens

L. Fontanesi, A. Ribani, V.J. Utzeri, G. Schiavo, S. Bovo
University of Bologna - Department of Agricultural and Food Sciences, Bologna, ITALY

Honey contains DNA from many different organisms that are part of hive micro-environmental niches and honey bee pathospheres. In this study we applied three approaches to identify honey bee pests, parasites and pathogens by analysing honey DNA: i) a targeted end-point polymerase chain reaction method with 18 primer pairs was used to detect DNA traces of a total of 10 pests, parasites and pathogens; ii) a targeted next generation sequencing (NGS) approach was designed to identify Varroa destructor lineages by analyzing eight regions of the mite mitochondrial DNA (mtDNA); iii) an NGS based shotgun metagenomic analysis was applied to identify all potential pests, parasites and pathogens. The two NGS approaches were based on customized bioinformatic pipelines for the interpretation of massive sequencing data. DNA extracted from a total of 110 honey samples (n. 77 from Italy and n. 33 from other European countries and continents) were analysed with the first approach. Presence of pests, parasites and pathogens was similar in honey sampled from all investigated macro-geographic areas: V. destructor, Melissococcus plutonius, *Galleria mellonella*, Nosema ceranae, *Lotmaria passim*, *Paenibacillus larvae* and Ascosphaera apis were identified in 99%, 81%, 60%, 57%, 50%, 47% and 40% of the analysed samples, respectively. Acarapis woodi, N. apis and Tropilaelaps spp, were not detected. The second approach was tested in 17 honey samples collected from all continents. All samples contained mite mtDNA of the K1 lineage. European and North American samples had also P1 mtDNA. The South American samples also contained sequences of the J1 mite lineage. A Chinese sample had mite mtDNA of the C2-1 lineage. The shotgun metagenomic approach was tested on two honey samples analysed with the other approaches. DNA from several other pathogens, in addition to those already detected with the targeted approaches, were detected including: fungi (Metarhizium spp., Bettsia alvei and Aspergillus spp.), bacteria (Spiroplama apis) and viruses (*Apis mellifera* filamentous virus). Honey environmental DNA, coupled with different analytical approaches, could provide valuable information for monitoring purposes simplifying surveillance against honey bee health treats derived by pests, parasites and pathogens.

The Impact of the Bee Informed Partnership (BIP) Technical Transfer Team

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The Bee Informed Partnership (BIP) Tech Transfer Team members work across the United States to bring commercial beekeepers, academic research institutions and industry together. The Tech Team members help support US commercial beekeepers, as they perform regular colony assessments and samplings, conduct industry trials and facilitate research experiments. Tech Team members support 110+ commercial beekeepers, who collectively manage approximately 17% of the total U.S. honey bee colonies. The Tech Team program efforts translates up to 32% fewer colony loss for the participating beekeepers compared to non-participants. In 2018 alone, they sampled over 10,000 colonies and collected data to contribute to the largest honey bee health database in North America. From observational studies to pragmatic trials and case studies, these highly trained field specialists bring a uniquely broad perspective to the U.S. beekeeping and honey bee research communities, and contribute to both test and share data-driven best management practices.

The Internet of Bees

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The internet of bees, or IoBee, is a project funded by the European Commission which aims to make major advances towards improved bee health. The diverse skill set of the IoBee partners promotes an interdisciplinary approach, which enables key innovative developments

such as closely monitoring activity at the entrances of bee hives, monitoring the density and diversity of wild pollinators in the field, big data collection and the processing and analysis of spatial information.
An optical bee counter has been developed to closely monitor activity at the entrance of the beehives, which gives valuable insights into the health status of the bee colony. The bee counter has been shown to identify the strength of the foraging force at any given time as well as mortality rates in field, deviations in flight duration and nectar availability. Furthermore, the bee counter can also identify pests entering and leaving the hive while classifying and distinguishing drone and worker bee traffic.
In addition, in field sensors capable of identifying both the density and diversity of wild pollinators as well as invasive insect species are being developed for application within apiaries as well as in crop settings and flower margins. Uses range from the establishment of sentinel networks to the generation of risk maps.
The project's aim is to facilitate the creation of a global network that includes data generated by the ground sensors, coupled with satellite data for vegetation, ground temperatures and other environmental variables to provide intelligent warning systems. These predictive tools will serve beekeepers and beekeeping associations to aid management decisions with respect to nectar flows, risks of infestations from pests such as small hive beetle and the Asian hornet, pathologies and the use of crop protection products. In addition, the data has the scope to inform public bodies and governmental authorities with decisions on pollinator conservation, land management and climate change.

Thermofluid analysis of the extended phenotype of a honey bee colony

D. Mitchell
Institute of Thermofluids, School of Mechanical Engineering, University of Leeds, Leeds, UNITED KINGDOM

Honey bees spend over 80% of their lifetime within the nest, and their very complex behaviours in finding, selecting and modifying it are well known, but to what end is all this behaviour exerted? How can one define metrics for the success or otherwise of the processes that utilise the extended phenotype? This presentation shows how the thermofluid characteristics of the honey bee nest bound the energetics of the colony and thus influence behaviour to determine outcomes throughout the year. It discusses the latest analyses of these key parameters in relation to: colony metabolic rates and metabolites; winter clustering ; nectar desiccation, fanning and foraging behaviours. It then shows how these parameters determine boundaries to minimum survivable temperatures in winter, the maximum range of foraging, viability of nectar sources, honey production rates, and the survival or suppression of varroa parasites. The analyses also provide insight on how the conflicting humidity and temperature requirements for the successful production of brood and honey are resolved. By these parameters providing easily accessible metrics for the fundamental differences between anthropogenic and natural nests, they become the criteria for improvement in the performance of man made hives and bee keeping practice in the face of adverse climates.

Heat power estimation of a bee colony in a Dadant-hive based on transient hygrothermal evolution

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Recent developments in electronic device (in terms of price and ease of use) have enabled us to equip beehives at low cost with sensors to monitor the hygrothermal behaviour in transient state of a colony living in a hive. It is nowadays easy to observe the already well-known fact that bees regulate their nest in temperature and humidity and have to continuously adapt to external varying climatic conditions for optimal development (in terms of health and production).
Our goal is to estimate the time-varying heat power produced by the colony (as a function) and to assess if it provides information about the colony health. This problem of function estimation is solved in two steps by inverse methods:
1) A thermal model of an empty hive is developed and implemented in Octave thanks to an empty hive equipped with sensors. While some sensors are part of model inputs (external conditions such as temperature, humidity, wind, luminosity), others (temperature inside the hive) allow us to tune the model parameters (related to materials and boundary conditions).
2) The thermal model is then extended to take into account the hive content (honeycomb, honey, bees). The comparison between the results given by the model for a populated hive and the real temperature measured inside gives an objective function (derived with the non-linear least squares method) which can be minimized to provide the time-varying heat power of the colony.
In addition, the thermal model of the hive can be used to simulate hives made out of different materials to evaluate their influence on the inner hygrothermal climate. Our results suggest that this factor could be more considered when buying or building hives than it is today. Furthermore, if it is confirmed that a consistent relation exists between heat production and the "health" of the colony, such results would enable beekeepers to monitor their hives without disturbing them (not opening of hive for example).

SYMPOSIUM OIE

11 SEPTEMBER 2019

SYMPOSIUM OIE I

13:00-15:00
ROOM 517A

OIE activities related to honey bee health

F. Diaz
World Organisation for Animal Health, Paris, FRANCE

The World Organisation for Animal Health (OIE) is an intergovernmental organisation established in 1924 with 182 Member Countries (as of March 2019). Its mandate is to improve animal health, veterinary public health and animal welfare world-wide; the health of bees is included in the OIE remit.
Under this general mandate, the OIE is dedicated to:
- ensuring transparency of the animal disease situation world-wide, including diseases transmissible to humans (see paper on the World Animal Health Information System),
- collecting, analysing and disseminating veterinary scientific information,
- providing expertise and promoting international solidarity for the control of animal diseases,
-guaranteeing the sanitary safety of world trade in animals and animal products,
- improving food safety from the farm to the abattoir,
- promoting animal welfare through a science-based approach,
- improving the legal framework and resources of national Veterinary Services.

In application of its mandate, the OIE has published different Standards related to bee diseases. They are mainly laid down in two publications: the Terrestrial Animal Health Code (Terrestrial Code) and the Manual of Diagnostic Tests and Vaccines for Terrestrial Animals (Terrestrial Manual).
For bee diseases, the OIE and its Member Countries can benefit from the support and expertise of several OIE Reference Centres.
The OIE also published recently a publication titled bee health and veterinarians (2014) providing useful information to all the actors involved in the management of the health of bees.

Biology of Vespa velutina in Asia

S. Martin
Salford University, Manchester, UNITED KINGDOM

The hornet Vespa velutina is one of the 23 species of hornets that evolved out of central Asia and now can be found throughout Asia and Europe. Although only one species, V. carbro has spread into Northern Europe. In Asia, V. velutina is one of the most widespread and cosmopolitan species ranging from the high mountains of China (People's Rep. of) to the dry lowlands of Flores in Indonesia, and consists of 12 colour forms. Its small size relative to other hornets means it is very agile and this allows it to become a predator of native Eastern honeybees (*Apis cerana*). However, *A. cerana* has evolved hornet evasion and warning behaviours, ensuring that it is difficult for the hornets to catch them. In 2003/4, V. velutina nigrithorax (known in Europe as the yellow-legged or Asian hornet) was accidentally introduced into both France and South Korea where they have now established large populations that continue to expand in both regions. This hornet species is well adapted to urban and semi-rural environments where it builds large nests, typically in the top of a high tree. The large nests are long lived (months) and harbour thousands of workers that require a large number of insects to feed the developing larva. In Europe, V. velutina encountered Western honeybees (*A. mellifera*) that lacked the behaviours to deal with the hawking hornets and soon were enduring heavy predation in the locality of hornet nests. Despite active research, the only current viable control method for beekeepers is the location and destruction (by experts) of nests. As V. velutina continues to expand its range in both Europe and Korea/Japan, the risks of a new accidental introduction into a new region of the world via the accidental transportation of a mated hibernating queen has increased. Vigilance is required as the impact on Western honeybees and other pollinators can be significant when V. velutina colonies reach high densities. Furthermore, it has been calculated that even if 95% of colonies in an area were destroyed (unrealistic) this would reduce the spread and nest density by only 50%, making control very difficult!

The yellow legged hornet Vespa velutina in Europe

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The yellow legged hornet Vespa velutina is an invasive insect predator that was accidentally introduced from east China into France around 2004. Since its arrival, V. velutina has expanded its range through Europe, impacting both biodiversity and beekeeping. In this presentation we overview the research themes that are currently being explored to understand and control V. velutina. After presenting the origin of the invasion, the expansion dynamic of Vv in Europe, and its development cycle, the different research teams and their research topics will be introduced. We next discuss the impact of Vv on the entomofauna, especially on honey bees, and describe its hunting behavior. Different control methods are summarized, and evidence presented for an urgent need for further research to detect nests as early as possible and to develop eco-friendly control methods. To better understand dispersal and foraging behavior, we studied Vv action range using a RFID system: its classic action range was under 1 km from its nest; less than half of the hornets could not return to their nest when released at further than 2 km from it, and the maximal homing capacity observed in our trial of 5km was only achieved by 4% of the hornets. New technologies are currently being studied to help detect nests: (1) Harmonic radar, allows users to observe insect flight trajectories with minimal disruption of their behavior, using a light passive tag, that re-emits microwaves transmitted from a min. 50 kg radar system. This system is cumbersome, and better adapted to open areas. (2) Radio-telemetry, is a well used approach to study animal movement but less so with insects owing to the weight of even the smallest active tags limiting their application to larger or stronger insects. The receiving system is light-weight, portable, relatively cheap, and can be readily used to follow insects through or around complex landscapes (e.g. with trees, buildings).The development of biocontrol solutions against Vv using entomopatogenic fungi is then finally discussed, after the discovery and study of several strains of fungi in France, able to parasite VV workers.

How is Vespa velutina nigrithorax dealt with in Korea?

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Department of Agricultural Biology, National Institute of Agricultural Science, Rural Development Agriculture, Republic of Korea

Vespa velutina nigrithorax is an aggressive predator of honeybees and other beneficial insects in ecosystem. V. velutina has been unintentionally introduced in Busan, Korea, in 2003 and is being widely spread across the country. As in the world, this hornet is considered as a serious pest of honeybees in Korea, as it preys upon domestic honeybees, *Apis mellifera* and *A. cerana*. Healthy beekeeping operations are seriously affected by this hornet predation. As a consequence, massive colony losses and damages are increasing vigorously. Currently in Korea, V. velutina is found in the majority of areas. Here we describe the control methods which are being used and newly developed against this hornet in Korea. The most commonly practiced control method is to directly kill the hornet using badminton pole. Conventionally, this hornet is physically controlled using a bait trap. Usually, in this bait trap sugar syrup or honeycomb are used to attract V. velutina. Another way is to spray pesticides or put hornet body in contact with them. In addition, we also recommend catching overwintering queen during spring season (April to June). Nevertheless, damages may persist. To overcome this problem, we attempted to find fundamental solutions such as (i) how to find V. velutina nest, and (ii) how to remove it. To achieve these goals, we integrated this control options with drone engineering technology known as an Unmanned Aerial Vehicle (UAV). UAV being a new technology, we focus the discussion on it and on engineering technology.

Criteria for the inclusion of diseases, infections and infestations in the OIE list

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The OIE list is a list of terrestrial and aquatic animal diseases, infections or infestations of compulsory notification by Member Countries to the OIE (181 as of 2017). Once a disease, infection or infestation is listed, the OIE: (I) verifies, validates and disseminates the information reported by each Member Country to other countries, which can take the necessary preventive action, and (II) can develop standards for harmonising disease detection, prevention and control, and for safe international trade in animals and their products. The OIE standards are recognised by the World Trade Organisation as reference international sanitary rules for animal health.

In order to be included in the list, a disease, infection or infestation has to fulfil 4 criteria. For terrestrial animals, the criteria are as follows:

1. International spread of the pathogenic agent has been proven.
2. At least one country has demonstrated freedom or impending freedom from the disease, infection or infestation in populations of susceptible animals.
3. Reliable means of detection and diagnosis exist, and a precise case definition is available so that an affected animal can be clearly identified and distinguished from other diseases, infections or infestations.
4. Significant impact on the health of either humans or domestic animals or wildlife.

The list is reviewed regularly by the OIE Specialist Commissions. Modifications (addition of new pathogenic agents or removal of diseases, infections or infestations already listed) can be proposed by Member Countries, experts from OIE Reference Laboratories and Collaborating Centres, International Organisations with which the OIE has cooperation agreements, or the Specialist Commissions themselves. If modifications are adopted by the annual World Assembly of Delegates (the highest authority of the OIE), the new list comes into force on 1 January of the following year.

For year 2019, the OIE list is comprised of 117 animal diseases, infections and infestations, including 6 that affect bees. These are: infection with Melissococcus plutonius (European foulbrood), infection with *Paenibacillus larvae* (American foulbrood), infestation with Acarapis woodi, infestation with Tropilaelaps spp., infestation with Varroa spp. (Varroosis) and infestation with Aethina tumida (Small hive beetle).

SYMPOSIUM OIE II 15:30-18:00 ROOM 517A

Adaptive Strategies of Wild Bees in Africa to deal with Threats

M. Lattorff
International Centre of Insect Physiology and Ecology (ICIPE), Nairobi, KENYA

Bees provide a valuable ecosystem service by pollinating plants and thereby contributing to nature conservation and food security. Managed and wild bees are threatened by a range of factors, which have led to declines of species during the past decades due to agricultural intensification and destruction of natural habitats. The declines of wild and managed bees are pronounced in the northern hemisphere. The southern hemisphere is less affected with almost no reports of declines of bee populations. This could be due to a true absence of declines or due to less accurate data. Indeed, a smaller percentage of African countries are reporting to OIE databases when compared to other countries. A lack of historical records makes estimates of declines even more difficult.
There is evidence that African bees are resistant, as they survive Varroa mite infections without treatment. This resistance might by due to large population sizes allowing selection to act efficiently and it might be enhanced by the high genetic diversity. In contrast to this, European and American bees have been selected for gentleness and high production reducing their diversity. Selection has also contributed to the establishment of large colonies providing rich resources for the establishment of pests and pathogens, while African colonies are much smaller. Behavioural mechanisms contributing to resistance of African bees might be the ability to detect and remove parasites more efficiently. In a comparison of American and African bees and their behaviour against the ectoparasitic mite Varroa destructor, African bees were more efficient damaging mites. African bees are absconding more frequently when disturbed, especially by brood diseases. Group-level defences acting against the intruders like small hive beetles by imprisoning them with propolis is well expressed in African, but not in American bees.

Tropilaelaps mites: news and impacts on honey bees

P. Chantawannakul
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Tropilaelaps mites are original parasites of the giant honey bees in South East Asia. Two of four species (Tropilaelaps clareae and Tropilaelaps mercedesae) are successfully adapted to parasite and reproduce in the European honey bee (*Apis mellifera*). T. mercedease are currently found parasitizing *A. mellifera* colonies in both tropical and temperate zones. Apart from human translocation, Tropilaelaps can be readily spread by natural causes such as robbing, swarming, and sharing food sources. By long evolution of host-parasite relationship, Tropilaelaps mites have not been reported to cause serious damage to Asian honey bees, due to their behavioral resistance mechanisms developed in Asian honey bees. Nevertheless, in *A. mellifera*, T. mercedease causes wing deformity, reduced weight and longevity, increased levels of bee viruses and eventually colony collapse. Global expansion is therefore being in great concern especially in the era of trade globalization and climate change that may favor the transmission, distribution, and ecological niche expansion of Tropilaelaps mites. We have adapted some techniques that have been previously applied to Varroa destructor to prevent and control Tropilaelaps mites in *A. mellifera* colonies, however, novel treatment strategies are needed for effective non-chemical control measures.

Virulent strains of known diseases

M. Schäfer
Friedrich-Loeffler-Institut, Federal Research Institute for Animal Health, Greifswald - Island Riems, GERMANY

Bacterial foulbrood diseases play an important role in honey bees and have shaped the apicultural industry around the world. American and European foulbrood are widely distributed, highly infectious and cause colony losses and considerable economic burden on apiculture. American foulbrood (AFB) is the most destructive and therefore the best studied bacterial disease of honey bees. However, different genotypes of the causative agent of AFB, the bacterium *Paenibacillus larvae*, exist. Of the four described P. larvae genotypes (ERIC I – IV), only ERIC I & II are frequently observed and differ significantly in virulence. The course of the disease on the larval level is more accelerated after infection with ERIC II strains allowing nurse bees to remove diseased larvae more efficiently before capping. For this reason the lead clinical symptom, conversion of capped larvae into 'ropy mass', is less frequently found than after infection with ERIC I strains bearing the risk of false negative diagnosis, a problem that is still insufficiently acknowledged and remains to be resolved.
European foulbrood (EFB), caused by Melissococcus plutonius, is also distributed worldwide and became an increasing problem in some areas, but many basic aspects of its pathogenesis and the disease epidemiology are still unknown. Anyway, using multilocus sequence typing (MLST) 34 sequence types (STs) were described that have further been grouped into three genetically distinct clonal complexes (CC3, CC12 & CC13). An evaluation of virulence under experimental conditions showed that an atypical strain of CC12 was the most virulent followed by a typical strain of CC3 and a strain of CC13 was not virulent. A large-scale field study in the UK largely confirmed the decreasing virulence from CC12 and CC3 to CC13.
Beekeeping practices and control-measurements have notable effects on the progress of foulbrood diseases and the beekeepers have

to understand their responsibilities to monitor and treat their colonies. Therefore, the competent authorities must disseminate adequate information on detection, diagnosis, control and management of foulbrood diseases.

Beneficial microorganisms and honeybees: colony level effects of lactic acid bacterial supplements

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Paenibacillus larvae, causative agent of the lethal American Foulbrood (AFB) disease, is the primary bacterial pathogen affecting honeybees and beekeeping. The main current methods for controlling AFB diseased colonies are either enforced incineration or prophylactic antibiotic treatment, neither of which is fully satisfactory. The search for superior means for controlling AFB has led to an increased interest in the natural relationships between the pathogenic and mutualistic microorganisms of the honeybee microbiome, and in particular the antagonistic effects of Honeybee-Specific Lactic Acid Bacteria (hbs-LAB) against honeybee pathogens including P. larvae. These effects have so far only been demonstrated on individual larvae in controlled laboratory bioassays. Here we investigated whether supplemental administration of these bacteria had a similar beneficial effect on P. larvae infection at the colony level in two different experimental set-ups.

First, we monitored treated and untreated colonies in AFB-affected and unaffected apiaries in Sweden throughout a season. The results showed that, over the entire season, the hbs-LAB supplements did not affect either colony-level hbs-LAB composition or P. larvae spore levels. Hbs-LAB composition was, however, more diverse in apiaries with a history of clinical AFB, although again this was unrelated to colony-level P. larvae spore levels.

A second colony-level experiment investigated whether supplemental administration of hbs-LAB had a beneficial effect on P. larvae infection by comparing experimentally AFB-infected colonies treated with bacterial supplements to untreated colonies and antibiotic (tylosin)-treated colonies, recording AFB symptoms, bacterial spore levels and various measures of colony health and performance. The results showed that tylosin mitigated AFB disease symptoms but did not affect P. larvae spore levels while the hbs-LAB supplements had no effect on AFB symptoms, P. larvae spore levels or colony strength.

These results do not contradict the antagonistic effects from hbs-LAB observed at the individual level but rather suggest that supplementary administration of live bacteria may not be the most effective way to harness such effects in a useful application.

Using bacteriophages against Paenibacillus larvae for the prevention and treatment of American Foulbrood disease in honeybee colonies

A.M. Alippi
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American Foulbrood (AFB) of honey bees is the most devastating bacterial disease of honeybee brood (*Apis mellifera* and *Apis cerana*) worldwide. Its etiological agent is the spore-forming, Gram-positive bacterium *Paenibacillus larvae*. Viable AFB spores remain in comb and woodenware for decades. Different methods of treating AFB-colonies are used, including the destruction of infected hives by burning and antibiotic treatments. Nevertheless, antibiotics are not currently recommended due to increasing resistance among bacterial populations and the presence of residual antibiotics on honey bee products. With the increasing demand for organic honey and the reduction of dependence on antibiotics, it is evident that an Integrated Pest Management (IPM) approach is needed to ensure the sustainability of the beekeeping industry. Within this context, the use of phage therapy seems promising and able to combine with an IPM strategy. Phage therapy is the therapeutic use of bacteriophages to kill bacterial cells by infecting and lysing bacteria. Since bacteriophages have shown high efficacy in treating bacterial infections in humans and animals, phage therapy seems to be an attractive alternative to control AFB. The main advantages and disadvantages of the use of bacteriophages and the prospects of AFB therapy will be discussed here.

BEEKEEPING ECONOMY

09 SEPTEMBER 2019

POSTER SESSION 01 08:30-18:00

BEEKEEPING ECONOMY I POSTER AREA

[P.01.01] Effect of the incorporation of information technology in hives to ensure traceability in honey production in Argentina

M.S. Vairolatti [1], G. Vairolatti [1], G. Cerutti [2], D. Gonzalez [2], J.P. Bono [2]

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The difficulty in identifing the honey origin and avoiding adulteration of it has increase the concern of the government, public and private organizations of the sector and the community, by virtue of the health hazards that this may cause. The purpose of this reserch is to identify the main efforts, in favor to offer best traceability in the productive value chain, to guarantee the aptitude of the honey arrives to the consumer, in accordance with the requirements of government agencies. For that reason,current status on the honey production was analized considering producers, collectors, exporters,public/private association of the sector and government.

As a result of the work, a model of productive traceability is proposed, which integrates the different links that make up the value chain, supported for the use of TIC (app, IOT, Sensors,software) from the origin to the end of the chain. This proposed model offers different actors of the chain access and generation of the relevant information from the use of sensors inside the hives to promote greater knowledge about the production process used, as well as having the visualization of the process as a contribution to the validation of what has been done in the different stages of the process. The use of the new technology, as guarantee of each step of the beekeeping chain, allows us an added value for the commercialization of the products genuine and natural, allowing to testify the denomination of origin of the same.

[P.01.02] Establishment of Identification Techniques for Taiwan Longan Honey

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Honey is a natural health product with high nutritive value. However, honey might be adulterated in various ways. The most common adulteration method is mixing different syrups during or after honey production. Instead of visiting the C4 plants, honey bees prefer to visit the C3 plants for nectars. Therefore, analysis of the difference between stable carbon isotope in C3 and C4 plant is a method to detect the honey adulteration. In this study, 117 awarded longan honey, 27 certified longan honey, 20 non- longan honey, 10 certified non-longan honey, 66 commercial honey and 9 commercial syrups from Taiwan, 54 longan honey from Thailand, 19 China honey were subjected to our analysis. The result showed by AOAC 998.12, 3 of Taiwan awarded longan honeys were adulterated, 1 of Taiwan certified longan honey was adulterated, 4 of Thailand longan honeys were adulterated, 8 of Taiwan non-longan honeys were adulterated and 30 of Taiwan commercial honeys were adulterated. In addition, the δ13 value of 3 commercially sugar syrup (C3, cassava) were -26.28, -25.63 and -25.16, respectively. We assumed that the C3 sugar syrup was added to the honey in production process, thus it could not be detected by the current technique. Alternatively, more detection methods should be created for cross-comparison. The 2-Acetylfuran-3-Glucopyranoside (AFPG) was used as an auxiliary standard of LC-MS/MS, the result showed that AFGP was not detected in any pure honey sample, the AFGP content in C3 syrup was 1.92, 0.22 and 0.45 ppm, respectively. Therefore, 5 commercial suspected adulterated honeys were selected for AFPG analysis, AFPG content were found in 2 of these samples, hence the two samples were considered as adulterated. Finally, we also analyzed the protein content of Taiwan longan honey and Thailand longan honey, the result showed that the protein content has a significant difference, and could be used to trace the origin identification of longan honey.

[P.01.03] Producing high quality honey in Argentinian sub tropic

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Honey production in Northern Argentina is an ancestral activity. In the last twenty years, the development and application of technological paths adapted to subtropical environment has impacted in the efficiency of the process increasing honey yields and complying with the highest quality standards. In this sense, the adopted technology proposes harvesting of sealed honey, ripened within the colony in order to obtain a genuine product that preserve all the natural substances that honeybees add during this process. The objective of this work is the evaluation of the adjustment of the technological trail proposed in order to be adapted to climatic variations in the region. This work was carried out in three apiaries of different beekeepers from El Galpón, which is located in the Gran Chaco Region, Argentina (25 ° 24 'South Latitude and 64 ° 39' West Longitude), at an altitude of 625 MSNM, with average temperature of 20.2 ° C (maximum January

34.6 ° C / minimum June 5 ° C) and 714 mm average precipitation. Dates of adding honey supers and honey yields of three different apiaries were compared during 2017-2018 and 2018-2019 seasons. The combs presented more than 90 % of the honey cells sealed when harvested. In the last date harvest, if combs with less than 90 % of sealed cells were detected they were removed from the process and the beekeeper was informed about the non-conformity percentage of combs in his lot in order to apply corrective measurements . Time period between super addition in the colonies and the first harvest was between 95 and 100 days for the first season and honey humidity percentages fluctuated between 16 and 17.3%. During second season honey was harvested after 71-92 days after super addition with moisture contents between 17.5 and 17.7%. Honey yields obtained were 28.5 - 30.5 Kg/hive and 24.3 - 41.3 Kg/hive for 2017-2018 and 2018-2019 respectively. These results indicate that the technological path implemented allows obtaining high quality honey in the Argentine subtropics with acceptable yields.

[P.01.04] Production and quality of honey of *Apis mellifera* in tropical environments of Perù

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Peru has three tropical apicultural regions, the Coast (CS) located on the western slope of the Andes up to 500 m altitude, there is no regular rainfall except for the northern CS with average rainfall of 200 mm per year, temperatures higher than 24 °C in northern CS and above 20 °C in central and southern CS. On the eastern slope of the Andes is the High Jungle (HJ) between 500 and 1500 m altitude with average rainfall of 1600 mm per year, average annual temperature between 22 and 26 °C; followed by the Lower Jungle (LJ) between 80 to 500 m of altitude with average rainfall of 2500 mm per year, average annual temperature of 31 °C. Flowering curves September-October in the CS with important blooms of carob (Prosopis pallida), avocado (Persea americana) and blueberry (Vaccinium myrtillus); July-August in HJ and August-September in LJ with important blooms of secondary forests (Inga sp., Vernonia sp., Cordia nodosa) and citrus fruits (Citrus sp.), which coincide with the harvest season of honey. The predominant type of *Apis mellifera* is the hybrid of subspecies of European origin (carnica and ligustica) with the presence of African genes (scutellata). The beehives are of the Langstroth type, circular honey extractors of four frames, with stainless steel harvest materials and the production technology used is technified more than 100 beehives with an average production of 18 kg per hive, less than 100 beehives with production average of 10 kg per hive, the time of operculate greater than 90% of 9 frames in time of flowering is 15 to 30 days in a full rise. In transhumant beehives from HJ to CS (for avocado pollination) the average production is 23 kg per hive. The marketing is done directly in the producer's house and in the markets of the region. The quality of honey obtained responds to the requirements of the food code with averages of 17 to 19% humidity, 3.78 pH units, 13.5 acidity, 7.5 mg HMF.

[P.01.05] Major, Trace and Toxic Elements of Honey Samples from Dinder Biosphere Reserve

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Dinder Biosphere Reserve (DBR) was established in 1935 and designated as reserve in 1979. It lies in the southeastern corner of Sudan against Ethiopia. DBR is classified in to three types of ecosystems: Maya'a "meadows", Dahara, "high land "and Reverine ecosystems.
The present study attempts to characterize honey produced in the three ecosystems based on elemental profiling. The aim of the study was to check environmental contamination and to provide a method for ecosystem identification.
Twelve, major, trace and toxic elements of 15 honey samples collected from DBR were analyzed by the atomic absorption technique.
Obtained results indicated that no toxic (Co, Cd, Cr, Pb) elements were detected, and K was the highest element in all samples, ranged 202.2 – 280.7 µg/g. Other elements were found in varying concentrations without significant difference between honey samples from the three ecosystems. However, only, copper was found to vary significantly (P less or equal 0.04) between samples. Cu ranged 1.2 – 5.9 micro gram per gram. This interesting result could be used in conjugation with statistics in the field of ecosystems identification in the future.

[P.01.06] Physicochemical properties of different florals sources honeys from Iran

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Honey is a supersaturated solution of sugars, which contains approximately 80% carbohydrates (40% fructose, 35% glucose, and 5% sucrose) and 20% water. Also, it contains more than 180 substances, like vitamins, amino acids, minerals, enzymes, organic acids and phenolic compounds [1]. The composition depends highly on the type of plants utilised by the bee as well as climatic conditions [2]. Honey samples that are available commercially differ in quality on account of various factors like seasons, packaging and processing conditions, geographical origin, storage period and plants sources. Therefore, comparing the physicochemical properties of honey with different plants sources is important to determination honey quality.

In this study, we investigated and compared the physicochemical properties of several honey samples (150) collected from different regions of Iran with different florals sources. Samples were analysed for moisture, ash, total acidity, diastase activity, hydroxymethylfurfural (HMF), proline and sucrose according to Iran's National Standards methods [3] and AOAC [4]. The results obtained showed that moisture, ash, total acidity, HMF and diastase activity in all samples were in accordance with Iran's National Standard limits. The mean value of sucrose was lower five in all samples except cedar and Citrus honey that values are 3.44-13.96 and 2.37-14.89 respectively. The mean value of proline was variety of 228-1280 ppm in different honey. Proline in all honey was higher 300 ppm except Citrus.
The purpose of this study was compared physicochemical quality of honey purchased in different plants sources of Iran.

BEEKEEPING ECONOMY
10 SEPTEMBER 2019

POSTER SESSION 07 08:30-18:00
BEEKEEPING ECONOMY II POSTER AREA

[P.07.143] Demographic and socio-economic influences of urban beekeeping

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Urban beekeeping is a rapidly growing phenomenon in many parts of the world. Using Calgary, Alberta, Canada, as a case study, we examined demographic and socio-economic influences on beekeeping as a hobby among members of a modern developed city. Calgary, with an urban area of 848 km^2, has a population 1.25 million. Land use, demography, and socio-economics are generally heterogeneous among the city's 198 neighbourhoods.
Urban beekeeping increased in Calgary ten-fold in the ten-year period 2008 to 2017. Such rapid growth is of interest to city regulators and ecologists. Regulators are primarily interested in public safety, as reflected in neighbour disputes, stinging incidence, honey bee cleansing flights, and swarm control. Ecologists express concern that rapid urban hive growth may affect native bees and the distribution of floral species preferentially pollinated by honey bees.
Within Calgary, 29 registered beekeepers managed 121 honey-bee colonies during 2008, but 10 years later, 317 beekeepers kept a total of 1208 honey-bee colonies. During the same period, commercial beekeeping in the province of Alberta experienced only a 30% increase (226,000 to 325,000) in managed honey bee colony count.
To identify factors that affect the growth and distribution of urban beekeeping in the Calgary, we examined its association with neighbourhood variation in mean income, education, immigration, unemployment rates, population demographics, housing, and other potential influencers. The analysis detected moderate positive correlations with neighbourhood settlement age as well as median age and education of community residents. Weak negative correlations involved population density, unemployment rate, and percentage of immigrant residents in each community.

[P.07.144] The Re-Introduction of Queen Rearing and Nuc Production Has More Than Doubled the Income Potential of Honey Producing Apiaries in the Cold Regions of North America

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Modern queen rearing was pioneered in N. America by a small group of beekeepers in the north central and northeastern U.S. states in the late 1800's. Every aspect of the production, harvest and shipping of queens in large numbers was developed by this group of pioneers, and has never been improved upon. These men also routinely overwintered small colonies (8 Langstroth equivalent combs or smaller) both indoors and outdoors.
After 1912, when the first successful shipments of package bees were sent from the southern U.S. to the northern, honey producing states, the production of queens and surplus bees moved to the south, and was almost completely abandoned by northern beekeepers. The pioneers were remembered as the fathers of modern queen rearing, but their practice of overwintering extra queens in nucleus colonies was forgotten, and eventually widely considered to be impossible.
When I first started raising my own queens in Vermont in the early 1980's, I knew of only one other person in the northeast U.S. doing something similar. In 1986 I re-discovered, by accident, that small nucleus colonies could overwinter here outdoors with minimal packing. By 1988, I was systematically producing and overwintering extra queens and nucleus colonies. As far as I know, this was the first time this had been done in the northern states since at least 1925.
Now there is a huge, unsatisfied demand for queens and nucleus colonies overwintered in the cold regions of the U.S., and a new industry beginning to form and supply that demand. From a beeyard location that used to produce 2000-3000 lbs. of honey annually, it is now possible to sell $25,000.00--$30,000.00 worth of queens and overwintered nucleus colonies. The best use of these newly recovered techniques however, is made by businesses that produce queen bees, nucleus colonies and honey. In this model, each of the three parts supports the others, total self-sufficiency is achieved, and income is stabilized throughout the year, and from one year to the next.

[P.07.145] Local economy development thru hive product diversification (Apicosmetic and Health) Kao Táxkat Company

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Kao Táxkat, Xalapa, MEXICO

Mexico is the 6th largest honey producer worldwide and 3rd in global honey exports. Despite exports reaching 90% of honey production, the per capita consumption is very low. Local producer revenues are too low. Five years ago, the diversification proposal of hive products had grown into six lines: food, honey, gourmet, "apicosmetic", "health" and teas.

This small company is located at the state of Veracruz, where the fields are characterized by microflorations, with an impressive biodiversity and a wide variety of climates. These conditions foster the creation of unique types of honey such as “marangola”, “chayote” “campanita”, “tarai”, “mulato”, “jonote”, “mozote”, “azahar de naranjo” (orange blossom), “mielatos”, “multifloral”, and “pimienta” among others. Usually this honey is homogenized for export, but we respect the blooms and the typified honey are saled here in Veracruz.
Kao Táxkat offers to the consumer a wide variety of typified honey, all year long. Some other products are made for beauty and health. All the products are made from raw materials taken from the hives, like honey, royal jelly, propolis, pollen and bee wax. As a complement to the company´s hives, local producers also obtain raw materials in the same region and Kao Táxkat takes this advantage to manufacture added value products.
The production and manufacturing processes do not derive from a chemical, pharma or cosmetic laboratory. From the initial development of the hives operation for the production of raw materials, Kao Táxkat has been inspired by the curative, regenerative, proteinic and nutritional properties of the hives products. And so, the development of added value products was set.
Kao Táxkat is a registered trade mark and meets all government and commercial requirements in Mexico. In addition, our products are traded in the main electronic market platform in Mexico.
In conclusion, Kao Táxkat covers the unique market for the local development thru diversification where the added value is at least 50% the value of the raw materials.

[P.07.146] BeePathNet – Enriching the Urban Jungle with Bees - From the City of Ljubljana BEE PATH to the BeePathNet network

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A holistic approach to urban beekeeping, already developed in the City of Ljubljana, will be transferred to five bee-friendly European cities, and the BeePathNet network will create favourable conditions for sustainable urban beekeeping. Through raising awareness of the importance of bees, and hand in hand with everybody living and acting in the cities, we are contributing to a cleaner and greener environment, and also to the preservation of natural resources and biodiversity. The BeePathNet network connects 6 EU Cities – Ljubljana, Slovenia, as the lead partner, Amarante, Portugal, Bydgoszcz, Poland, Cesena, Italia, Hegyvídek - XII. district of Budapest, Hungary and Nea Propontida, Greece. Key project activities:Development of action plans on urban beekeeping in each partner city, Ljubljana BEE PATH good practice study tour with training for Transfer city ULG members, transfer-city Case-study and topic devoted partnership meetings on the maintenance of biodiversity in urban areas, BeePathNet educational packages, awareness raising, development of “bee paths”, and development of new bee products, development of a set of thematic guidebooks, newsletters and project booklet in order to transfer knowledge to stakeholders within the partnership and beyond. BeePathNet presents a platform for long-term activities in partner cities and wider as well as development of different opportunities for beekeepers in the cities. This will enable them to upgrade their activities and develop new products, what will empower their economic status and make them more visible in the community. Such a partnership composition allows the BeePathNet project to cover the majority of climate conditions for beekeeping in the EU (Atlantic, Continental, Mediterranean, Alpine), as well as to take into account cultural and social differences between the cities. Additionally, partners were selected, based on their specific knowledge and good practices, which will be their contribution to the transfer process.

[P.07.147] Industrial Internet promotes the sustainable development of China’s bee industry

Z. Zhang, L. Liu
Hangzhou Smart Beemap Network Technology Co., Ltd., Hangzhou, CHINA

China leads the world in the digitalization of the front-end consumer side. Consumer behavior is highly digitalized, and digital innovative applications and business models are emerging.On the contrary, industrial Internet is still in the development stage on the whole.For example, the digital application and development of bee industry, which is produced by traditional small farmers with highly dispersed production groups (beekeepers), is still in a backward stage.Based on the analysis of the existing problems and pain points of traditional beekeeping industry in China, combined with the characteristics of beekeeping industry and Internet platform services, this paper introduces the innovative development of bee industry Internet in China through the Dr. Bee platform developed through the digital application of the whole industry chain before, during and after the beekeeping industry.We provide remote technical exchange and training for beekeepers in the community on how to use the Internet technology of the bee industry.Digital cloud apiary management,Internet of things intelligent bee machine development,Application of blockchain technology in the source traceability system of bee products,The development and application of bee pollination trading platform and bee product supply chain management and other aspects are explored in the digital application, in order to seek for the sustainable development of China’s bee industry in the future and efficient service of agriculture solutions.

[P.07.148] The impact of beekeeping education to reach horizontal educational objectives

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Grm Novo Mesto - The Centre of Biotechnic And Tourism, Novo Mesto, SLOVENIA

Today's education in the field of beekeeping in Slovenia based on the rich heritage of beekeeping and has today an exceptional dimension. Bees, beekeeping and the life of wild pollinators are known all over Slovenia, from children, students of all generations, as well as adults. They are faced with beekeeping technology issues, great importance is also given to understanding the multifunctionality of the beekeeping area, such as getting known of beekeeping cultural heritage, developing apitourism, apitherapy, honey culinary as well as developing food products based on bee products and contributes to environmental awareness and other benefits of agriculture.
There are 11 schools in Slovenia that educate young people aged 15 to 22 years and adults in various andragogical programs in the fields of agriculture, food processing, horticulture, forestry and nature protection and are grouped together in the Consortium of Biotechnical Schools of Slovenia (CBSS), in which students also meet beekeeping issues. Most of these schools also have the appropriate infrastructure for practical work in beekeeping.
We made a research among students and lecturers CBSS in order to find out in which subjects students are studying beekeeping and to what extent beekeeping themes are used to achieve horizontal educational goals such as understanding the concept of sustainable development, the goals of conserving biodiversity, development green tourism and other general and specific educational objectives.
Using the method of surveying and interviewing, we assessed the achievements of integrating beekeeping topics into education in the field of horizontal education goals, harmonized with international strategies, and proposed a model for the optimal integration of beekeeping contents for integrated agricultural and nature protection education.
The results of the research show that the integration of beekeeping content into the educational process at different levels of education and training covered horizontal objectives, in particular in the field of education for sustainable development and the conservation of biodiversity, which contribute to a general increase in environmental awareness and the development of green jobs.
A high level of knowledge of beekeeping in the wider population has a beneficial effect on the economic result of the beekeeping industry.

[P.07.149] Building resilience through integration of honeybees in large cardamom farm, Nepal

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International Centre for Integrated Mountain Development, Kathmandu, NEPAL

Large cardamom (Amomum subulatum Roxb.) is one of the oldest indigenous spices of the eastern Himalayas. It is a boon for mountain farmers because it is a low-volume, high-value and non-perishable product. It is a micro-climate specific crop and found mainly in eastern Himalayan region with comparative advantages for positioning in the market with Geographical Indication (GI). Nepal is the world's largest producer, with a total of 6,439 metric ton of cardamom, followed by Sikkim, India with 4300 metric ton, and Bhutan with 2,091 metric ton. ICIMOD study shows that the large cardamom contributed US $500-1,700 to a households' annual income in 2014.
In recent years, extreme climate events such as drought, erratic rainfall, unpredictable hailstorms and snowfall have begun impacting yield as well as the crop cycle. At the same time, the increased dependency of farmers on cardamom as a primary cash crop poses higher risks because of production instability and price variability. We have therefore carried out action research and pilot projects to develop a package of practices and showcase interventions for reducing risks of production and market fluctuation through diversification of income sources. Integration of *Apis cerana* beekeeping, legume crops and kiwi fruits is one of the recommended actions implemented in the project site in Nepal.
The baseline and endline evaluation revealed that the communities who applied package of practices for water, soil/nutrient and crop management as well as a recommended business model were less affected by the price fluctuation and yield variability than others. The findings also revealed that fruit set, seed set, and fruit and seed weight (capsule quality) were all significantly higher in plots with *Apis cerana* colonies than in plots without a bee colony.
This paper highlights the risks and vulnerabilities in large cardamom farming and share the overall strategies for building resilience of farmers and entrepreneurs involved in and around large cardamom value chain, including recommendation to integrate *Apis cerana* beekeeping.

[P.07.150] Beeswax yield quantification obtained from different crude beeswax sources for commercialization in South Western Oromia, Ethiopia

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Ethiopia is known for the availability of large amount of crude beeswax as raw materials for pure beeswax. But the information on the amount of recovery of pure beeswax is not well documented. The objective of this study was to determine the amount and quality of pure beeswax recovered from crude beeswax resources. The existing sources show that crude beeswax is obtained from honey wine making houses residues, honey combs and uncapping during honey extraction. Due to the absence of enough information on the recovery

percentage of pure beeswax on recovery percentage is causing a serious controversy between tax authority and processors companies. This factor seriously affected export of beeswax which in term affected producers in particular and the nation in general. To solve the controversy, samples were collected from 17 representative sites and bulked into six homogenized samples for pure wax extraction. The result show that the amount of pure wax recovered ranges from 18 % and 32.2 % with effective mean of 28.4 %. Therefore the processors companies extraction of crude beeswax could be save from seek low cost and efficient crude beeswax refining technologies that can be affordable to beekeepers. Further studies on identification of the optimum temperature and time required in heating crude beeswax for extraction process and conducting research review to investigate the crud wax wastage due to mishandling at local producers.

[P.07.151] Use of bee products to strengthen health of people: experience and achievements of Ukraine

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Private Business, Brand Medok, Kharkiv, UKRAINE

Now over 400 thousand beekeepers are working in Ukraine and they produce not only raw honey, but also various bee products (pollen, bee bread, propolis, royal jelly, drone's homogenate, etc.). Along with raw home made bee products, Ukraine produces and sell certified processed bee products which can be used in apitherapy and pharmacy in Ukraine and worldwide.

Some bee products have received official status of functional healthy food and recommended for prevention and mitigation of various diseases in Ukraine. In addition to guarantee of food safety, modern processed Ukrainian bee products have an attractive design, modern and convenient packaging, and labels with detailed information about its properties and method of application.

Currently eight categories of bee products are represented at market in Ukraine: 1) Propolis: tinctures, extracts, sprays, honey, candies, dental elixir, suppositories; 2) Royal jelly: dry in capsules, tablets, suppositories; 3) Homogenated drone's larvae: dry in capsules and suppositories; 4) Dry dead bee bodies: dry powders, tinctures, extracts, ointments, gels; 5) Pollen and bee bread; 6) Tincture of wax moth larvae (Galleria melonella); 7) Dissected comb's caps (zabrus); 8) Wax. Some of bee products are new, original, unique, recently patented and produced only in Ukraine.

It nice to emphasized that bee products are well demanded at market in Ukraine. At the first, Ukrainian production of bee products provides competitive prices which begin to successfully compete with pharmaceuticals. And secondly, the Ukrainian beekeeping enterprises are very actively engaged in the popularization of bee products and the promotion of apitherapy. Main management task is to transfer bee products from exotic or niche sales into daily products which everyone use regularly.

Now in Ukraine bee products are sold at beekeeping fairs, beekeeping shops, markets, healthy and organic food stores, phyto-pharmacies, etc. Retail market is represented by off-line and on-line stores. In order to increase consumption, the strategy of promoting bee products in Ukraine pays great attention to the health of people, including disease prevention, strengthening human immunity and improving the quality and longevity of people.

[P.07.152] Apiforestry to replace charchoal burning

J.P. Komakech
Jing Komi Farmers Groups, Gulu, UGANDA

Replacing Charcoal Business with Apiforestry"is an alternative livelihood campaign for this project to be in case an interested partner donor is identified for funding against the current indiscriminate cutting down of trees for charcoal to earn livelihood in Uganda by promoting fast growing fruit trees/crops with good nectar sources in addition to conservation and planting of the native tree species to boost honey productivity and to diversify income sources for beekeepers since the forage have diminished due to charcoal business instead of honey business and thus "Apiforestry". If funded the project will be implemented with bee farmers in GULU and OMORO districts, Northern Uganda. The project seeks to engage unemployed youth, women and smallholder farmers in the rural communities through Integrated Beekeeping Model to diversify income with growing of passion fruits for short term but for long term the project will be planting crops like mangoes, avocados, oranges, limes, guavas, papaw, Shea-nuts for income generation and nectar sources. The project seek to support 500 beekeepers 75% youth (men & women) through training in modern beekeeping practices, provide technical support through mentorship to set up suitable apiary for quality honey production, supplied with manufactured modern hives (KTB) and necessary equipment for high quality honey harvest. Out of the 150 beneficiaries, the project will select 25 youth (men & women) train and mentor them to become product processors for value addition like making of honey wine making (from honey and fruits), Candle making, shoe polish, propolis tincture, Vaseline, Soap, mead, tooth paste so for them to become model youth to inspire more youth to engage in beekeeping as a mean of employment and sustainable livelihood.

[P.07.153] Beekeeping sector in Lebanon – State of Art

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Beekeeping in Lebanon for a long time was considered complementary activity for farmers and a hobby for others. And because of its importance the Lebanese Agriculture Research Institute has founded a Unit for beekeeping.

In 2018 and as a part of its activities under Sustainable Development Goals 1, 2, 11, 12 and 15 the unit has launched a new and comprehensive assessment targeting the totality of the beekeepers in the sector. The assessment focused on Honey and other hive products Honey quality origin and chemical and floral properties were also checked. Questions related to bee diseases were also asked. Focus groups for the different stakeholders of the sectors were scheduled. The unit also established a beehives monitoring systems in addition to capacity building for ministry of Agriculture staff (trainers of beekeepers).
At the end of the assessment a baseline survey report for beekeepers and pests and diseases was published. A thorough discussion of the results was made and highlighting the strength and the opportunities of the sector and the strategy for the sector for the coming decade was outlined.

[P.07.154] Creativity & Diversification of offerings equals profitability plus educates and inspires your customers to care for pollinators

L. Stahl
Beebees All Naturals, Brownstown, USA

With people flocking to beekeeping beekeepers who expand their product offerings beyond the typical honey & beeswax products will stand out in the marketplace, generate repeat customers and create more profit. This talk will touch on income generating possibilities for beekeepers. Bee & Swarm Removal, Mentoring, Education (camps for children, educational events for groups and retirement homes & Workshops (such as tea and honey pairing talk, varietal honey tasting, beeswax candle making, lip balm making, etc.) Honey & very creative and unusual specialty honey, such as Bourbon barrel aged, cold smoked over cherry wood, Tumeric and black pepper & some seasonal offerings such as mint for Summer & chocolate orange for the holidays Honey Lollipops, old-fashioned clear toy candy Salted Caramels & other candy. Soaps made with beeswax and honey made in bee themed molds & other bath products. A variety of creative and interesting things to create from beeswax including candles creatively packaged, beeswax birthday candles, also fire starters, ways to sell wax, wood treatment & paw wax to protect pet paws. Art objects such as handcarved wooden honey differs and paintings with honeybees and inspiring messages. We will also touch on the value of collaborations with restaurants chefs bartenders, Brewers Mead makers and distilleries. Also discussed will be possible medicinal avenues including bee venom therapy, propolis tincture kits, salve, propolis diffusers & other products such as pollen, royal jelly, and propolis toothpaste and gum.

[P.07.155] Diversifying income sources for beekeepers

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Organisation pour Promotion des Arts apicole et Sylvicole, Niamtogou, TOGO

Income, in beekeeping, is basically derived from goods such as pollen, honey, bee-bread, larvae, royal jelly, wax, propolis, venom; or services as therapeutic sting and pollination. Other good business opportunities are also found, as demonstrated in the following paragraphs, inside the main parameters of production that are the apiary, the harvest workshop and the beekeeper. The apiary, this ecosystem of space usually poorly wooded, with a fauna dominated by bee, main reason of enormous concentration of lizards, hedgehogs and sporadic presence of snakes, gives two categories of business. First one the exploitation of the surrounding soil which one can reorganize for a part in dense forest with well-chosen species and other one in plots of seasonal crops. The second is the creation of wealth from emigrant vertebrates: on the one hand the assembly of a small rearing of small predators to feed with the lizards and on the other hand a win-win research project in-situ in partnership with zoology institute on the biology and manners of hedgehogs and of snake. From the harvest workshop, the following income-generating activities can be glimpsed: the production of sweetened or alcoholic beverages from residual honey and second-choice honey; the feeding strengthening of small poultry from the oil-extracting cakes of wax and honey from the old rays of brood and pollen ; the manufacture of hollow concrete buildings, useable depending on shape and dimensions, as beehive, washbasin, water tower, pot both for hydroponic cultivation, trees feeding, or for wooden poles pesticide treatments, by a mould from our invention whose master component is in wax. The beekeeper, basically, meticulously observes every times the flora state in relation with the meteorological parameters. He unexpectedly discovers precious species development and germination conditions; some wild animals in their manners. Thus he becomes a data bank useful for the preservation of the biodiversity marketable or for self-one use. The struggle of bee companies to explore in full the basic sources of income as well in their variants as in their various sub-opportunities is an ideal for diversify taking means of beekeeper. Diversifying beekeeper's income sources appears require qualifications and ingenuity.

[P.07.156] Queen Breeding in Canada: An Overview

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Canadian beekeepers import over 250,000 queen bees from the U.S., Chile and New Zealand each year. These importations present significant risks to the Canadian beekeeping industry, including the importation of pests, pathogens and undesirable genetics (including Africanized bees), as well as stock poorly-adapted to our northern climate. In addition, Canada has a precarious dependency on foreign

sources of bees. In the not unlikely event of a temporary or permanent prohibition of foreign queen importation, the resulting agricultural and economic impact to Canadians would be negative and severe.
Over the past four years, through the BeeOmics project, our team has surveyed over 150 Canadian beekeepers and bee breeders from across the country, as well as provincial apiculturalists and technical transfer team members, to gain a deeper understanding of our current queen breeding industry and the opportunities and challenges that lie ahead. This presentation will highlight key findings of our data collection including importation and colony mortality data, beekeepers' perspectives on breeding, as well as best practices for moving forward. We also show the value of adopting new genomic breeding tools into breeding operations in Canada to create stronger, more disease-resistant stock (Canadian Honey bee queen bee breeding guide 2018, https://honeycouncil.ca/honey-industry/canadian-honey-and-honey-bee-stock/).
Our overall findings reveal an optimistic industry with great potential for a sustainable future of breeding high-performing Canadian-selected queens and colonies while minimizing the risks associated with mass importations. These findings are derived from Canadian data, however, the value of expanding a domestic breeding industry is universal, and many of our best practice recommendations would be relevant for other beekeeping nations.

[P.07.157] Comparative study on the adoption of improved beekeeping technology for poverty alleviation

Y. Ghatane Sunar
Shree Mahalaxmi International, Butwal, NEPAL

Adoption of improved beekeeping practices was compared between the mobilized (Pragatinagar VDC) and non-mobilized (Makar VDC) farmers' groups (n = 14 in each VDC) of Nawalparasi district using semi structured questionnaire survey after introducing improved beekeeping practices during 2002/2003. Majority of the households (82.4%) from mobilized group practiced beekeeping enterprise of which 80.6% followed improved practices with adoption index of 77.44% while from non-mobilized group only 56.0% adopted beekeeping enterprise and 68.4% followed improved beekeeping practices only with the adoption index of 58.73%. Annual honey yield per colony was significantly higher among mobilized farmers' group (25.6 kg earning NRs 25,657.14) than that of non-mobilized farmers' group (15.6 kg earning NRs 10,364.29). Women
involvement was low in enterprise development and adoption in both VDCs (31.8% and 50.0% in mobilized VDC; and 35.7% and 48.2% in non-mobilized VDC). Therefore, transfer of improved technology to subsistence farmers emphasizing women through social mobilization could help generate income and alleviate poverty.

[P.07.158] Diversifying income source for beekeepers. Propolis production in Uruguay

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In Uruguay, most of the beekeepers are small or medium producers, who develop their activity in a family way. As a complement to the production of honey, propolis is produced and sold to local laboratories or exported naturally.
The production is done by scraping materials from the hive in disuse, or using plastic mesh that is placed under the roof of the hive. This last technique is chosen by laboratories, given the decrease in impurities in propolis. During the year 2017 to 2019, technical and monetary support was provided by the MGAP (PPIR Program, More Inclusion for Rural Development), for investment in plastic nets of collection of propolis and to coordinate the sale to local laboratories. The meshes were placed in the hives avoiding contamination of sanitary products and the yields were analyzed. And the materials were scraped with a sharp element, to obtain the impregnated propolis. A study of the quality of the scraping propolis was also carried out (in charge of the laboratory), and it was related to the geographical origin.
The joint sale of propolis (18 beekeepers) is currently 640kg total. All beekeepers today handle the meshes in spring and summer. A hive can yield two meshes per year depending on the vegetation surrounding the apiary. We estimate that the apiaries with the highest yield in the hives are those that possess diversity of Baccharis sp. and native forests in their environment (up to 160gs per hive in the meshes). While prairie environments and Eucalyptus sp. Forests are the lowest performance (Up to 70gs per hive).
Regarding the quality of the scraped propolis, the following data were obtained (min and max): resins(51.1-74.3%), wax(9.3-33.8%), residues(6.25-15) , 69%) and flavonoids(107.1-172.9 mg flavonoids / gr PEP-100). The group work allowed the exchange of knowledge between beekeepers and the joint sale improved the price of the product.

[P.07.159] Beekeeping and Honey Hunting in Nepal: Current Status and Future Perspectives

S.K. Dhungana
Tikaram Foundation, Kathmandu, NEPAL, [2] *New Ashok Enterprises, Kathmandu, NEPAL*

Nepal is rich in honey bee diversity. Four native species of honey bees viz. Apis laboriosa, *Apis dorsata*, *Apis florea*, and *Apis cerana* and one exotic species, *Apis mellifera* are found in Nepal. Beekeeping with *A. cerana* was started in 1960, whereas *A. mellifera* in 1990. The Nepal Government and several foreign agencies have been involved in promoting beekeeping for the development of livelihood of deprived communities in rural areas, but beekeeping has been hindered due to lack of modern beekeeping technology, improper management of colonies, and lack of techniques in controlling mites, and brood diseases. The Nepal government and hundreds of international NGOs/INGOs are distributing bee colonies, honey buckets, and some hive tools to villagers for poverty alleviation without any proper beekeeping training. In addition, honey hunting from cliffs is a spectacular event for tourists to enhance the income generation of mountain people. Due to deforestation, and over-harvesting, the cliff-nesting species, A. laboriosa, is in an alarming decline. Other Apis species and 34 species of bumble bees have been recorded in Nepal, pollinating wild plants, as well as some of the cultivated crops.

[P.07.160] Himalayan Honeybees and Beekeeping in Nepal

S. Bishwokarma [1], N.B. Bishwakarma [2]

[1] *Sunil International Trading, Kathmandu, NEPAL,* [2] *Nepal Disabled and Helpless Rehabilitation Center (NDHRC), Kathmandu, NEPAL*

Nepal, the central Himalayan Republic, has five geographical regions: High Himalaya, High Mountain, Middle Mountain, Swanlike and Terai. There exist four native species of honeybees, Apis laboriosa, *Apis dorsata*, *Apis cerana* and *Apis florea* are found fromTerai up to the base of Himalaya. A cerana are kept in Traditional log and wall hives. A cerana is very aggressive, frequently swarms, and easily absconds, but is well adapted to the extremely cold climate conditions of Himalaya. *A. cerana* usually swarms two times: in summer (March-May) and in winter (November-December). Subsequently honey is also harvested in summer and in autumn. Beekeeping with *A. cerana* means not only of income generation for traditional beekeepers, but also a valuable resource of Himalayan regions.

Conclusion

The beekeeping with *Apis cerana*. was started in mid 19960. Agriculture department and several foreign aid agencies – UNICEF, IUCN, ICIMOD, MEDEP (UNDP) – have been involved to conserve the Himalayan honeybee, *Apis cerana*, and to promote beekeeping for more than two decades, but the number of colonies persistence and honey yield data reveal that *Apis cerana* beekeeping is hinder due to cyclic occurrences of sac brood diseases and lack of a proper knowledge of modern beekeeping. On the other hand, the wild bee, Apis laboriosa, is a rare species still exploit for honey, brood and wax production. If the nests exploitation by human predators cannot stop, than Apis laboriosa will disappear forever from the whole Himalayan ecosystems. So it is urgently need to conserve this species to maintain the biodiversity of Himalayan regions.

BEEKEEPING ECONOMY

11 SEPTEMBER 2019

POSTER SESSION 13 08:30-18:00
BEEKEEPING ECONOMY III POSTER AREA

[P.13.296] State and challenges of the honey market and the beekeeping community in Mexico

F. Berron
Amema, A.C., Merida, MEXICO, [2] Miel Integradora, S.A. De C.V., Merida, MEXICO

Mexico is one of the 5 Mega diverse countries in the world, thus its honey industry has suffered many negative impacts from the one analysis approach to honey authenticity tests.
False positive results to adulteration LC-EAIRMS C3/C4 analysis have occurred because honey blends of diverse botanic origin had pure honey contributions in the opposite spectra of the C3 isotopic values. These findings lead to the understanding of the use of the test as benchmark to “construct” fraudulent honey with C3 syrups. Syrups that could acquire the name of honey in such self-confidence to pay the honey import duty in Europe or the US and cross borders as “animal products” which they are not.
Another history of false positives, have been marker tests like SMB (never made available to the public) were developed and offered to importers, without acknowledging the local biology of honey producing countries, causing many problems and economical losses. Later in time responsable laboratory dropped the SMB analysis, upon realizing they were wrongly tagging as adulterated perfectly pure honey.
Rules of quality are seldom implemented to favor tropical producers and their honey is severely punished if HMF will arrive to honey packers higher tan 10 to 15 PPM whilst the limit for Tropical honeys in European and world regulations is 80 PPM.
Short videos are presented of indigenous small beekeeper families and of commercial beekeepers explaining the impacts of unsold honey in hot tropical places where HMF develops surprisingly fast and lethal as price killer. These are some Latin American impact stories of the honey fraud unveiled in the UK, USA, Australia, Spain and France where evidence of massive fraud is undeniable.

[P.13.297] New Markets and Opportunities for Apiculture in India

S. Rao
Honey R Us, Business Firm, Bangalore, INDIA

India’s market is better poised for innovative products and services related to this field. The talk will cover the following aspects; Culture, Consumer, and Commerce, the three C’s, with respect to Apiculture (Honey, Honey Products, and Api-therapy). In other words, the talk will touch upon the under-exploited opportunities.
Culture: India offers a rather unique opportunity as a result of the grasp of ancient civilizational medicinal practices and the resurrection of Ayurveda. A significant part of the population understands that food and nature offer clues, prevention, and cures to man-made ailments. While the West has surged ahead in understanding the science and practice of Api-therapy, its adoption has not been as high as desired.
Consumer: While India is a large producer and consumer of honey, the understanding of the composition of Honey and its properties are still at its nascent stages while the belief about their goodness and effectiveness remains strong. Though honey is the only product mostly consumed in India, there is a larger market for more innovative honey based products.
Commerce: India plays a dual role as both a consuming and an exporting nation to its neighbors to the East. The “Make in India” opportunity propelled by Government of India holds many opportunities for the Apiculture fraternity across the world. So far, the world has seen India as a consuming nation but not so much as a value-adding nation. Early mover advantage exists. Companies and countries have experimented with produce in India for sale outside of India in the agriculture space. Many non-agriculture companies are also establishing their presence to produce in India for both domestic and international sale. Incentives are attractive.
In conclusion, International Apicultural Congress is a vast amalgam of brilliant scientists, practitioners, business people of these angels of agriculture, ecology, and life. The funneling of the cumulative experiences available is remarkable and the coming together of such experiences in India is both commercially and socially very compelling.

[P.13.298] Modern management on raising international sales of honey and its products

V. Salehnezhad, S. Saleh Nezhad, A. Ziaei Boukani
Shahd Golha Co, Mashhad, IRAN

In this paper, we have tried to examine the impact of modern management and modern marketing methods on the international sale of honey and honey products. Many beekeepers cannot use costly marketing and advertising methods to introduce their company and their products. Accordingly, the use of low cost and new methods for marketing managers of these businesses is a vital necessity. In this article, we have tried to introduce low-cost methods that can be useful for any director in the exporting companies of honey and all beekeepers which are mentioned below.
1. Know your customers: Describe someone who is most likely to want or need your product. Why should you buy your product, when you

understand the motives, you can direct the product to the right customers? You cannot sell a product unless it is defined and positioned.
2. Count the product's properties to the client inside or outside of the country in a clear and elegant manner: All they need is to show them what will be happened for them when they use your products, and wait for a growing sales miracle.
3. Demographics know the sales areas around you: Demography is the area in which you live or want to present your product. How are income levels?How much do people would spend on your honey production? How are they religious beliefs and religious tendencies about your product? If this area does not have the capacity to buy your products or they will not use your products, then you have been dropped out of business even before you start working and before it will be operational.
4-Common Marketing: If you find that you are not alone in bearing the cost of marketing, look for a partner whose product is appropriate for your customer, or that your products complement each other. By offering a marketing plan, you can save on marketing costs.
Generally, there are many creative ideas for promoting honey business, it is important to spend time and effort on developing a coherent marketing and management program that outlines the main ideas for marketing and selling honey.

[P.13.299] Honey marketing for small producers

B. Imani, J. Dolabi
Asal Malake Dolab, Sanandaj, IRAN

Bee's productions are different throughout the world, due to variety in flower types, vegetation, etc., In terms of taste, aroma, color and usefulness. In order to all people of the world can access the resources, also the products in vast areas not be exclusive to a single person or company, and the smallest bees be able to sell their products to the world and sell at the best prices, it requires the launch of a global site by executive directors like APIMONDIA.
The site should be simple, strong in terms of the introduction and support of beekeepers and also be robust in terms of enforcing the rules.
The process of forming this site should be as follows:
All countries have the chance to be present once they are verified by the local cooperative of the area. Ultimately, the approval of the list of provincial cooperatives by the national union and its registration on the site system should be comprehensive and complete.
The beekeeper provides the following complete information to APIMONDIA;
Full description of the beekeeper
Deployment area
Vegetative flower type in the apiary area
The type of bee feeding
Used drugs
The analysis of the honey produced with the photographs or documentary films set up by the beekeeper, Of course with approval by the cooperative and the union to post on the site:

1. Registration of the country and the data about areas of deployment
2. Evaluation of each country or region
3. Competition between countries and the union and cooperatives
4. The system of sending and receiving the product
5. The annually payment of cost of maintaining, promoting and introducing the site
6. Each beekeeper will be removed of the site if a violation of the system occurs
7. Compensation to the consumer by the union of the country concerned
8. exchange of information of beekeepers

There are other ways to manage the site best which will be submitted to the Congress in more detail if the article is selected.

BEEKEEPING ECONOMY

12 SEPTEMBER 2019

POSTER SESSION 18 08:30-18:00

BEEKEEPING ECONOMY IV POSTER AREA

[P.18.434] Honey value chain analysis in Godere District, Gambella Regional State, Ethiopia

G.L. Yadeta

Oromia Agricultural Research Institute, Holeta Bee Research Center, Holeta, ETHIOPIA

Godere is one of districts in Majang zone of Gambella Regional State. About 40% of the district is covered with dense forest called Godere forest covering about 120,000 hectares of land. The flowering calendar of the major honeybee plants indicate that there are three honey production cycles. The major honey harvesting season is April-May, after the honey plant Scheffleriaabyssinica flowers. This is followed by another honey flow season in June, from honey plant Croton macrostachyus.Moreover, honey plants like Guizotiascabra and Bidenspychlomaproduce significant amount of multi-floral honey during November-December. The average crude honey yield per traditional hive per year is about 27kg, that gives the total crude honey about 583MT.The honey value chain is typical of the traditional honey production and marketing: no improved beekeeping input suppliers in the nearby, very poor extension services, almost no value addition and poor marketing The primary actors in honey value chain in Goderedistrict are smallholding honey producer beekeepers, honey collectors, mead (tej) makers, honey retailers and consumers.The market is characterized by long-term business relationships, trust and personal connections between producers and local honey collectors and to some extent between beekeepers and tej producers. The major share of honey marketed goes through the intermediate buyers (honey collectors and traders)who appear to the nearest village markets during harvest season.Tej makers play significant role in honey value chainthrough creating demand for the honey and processing and transforming the crude beeswax.Generally, the honey value chain analysis of the area indicates that almost all the produced leaves the district with no value addition, only passing through the longstanding market channels. Therefore, in order to utilize the huge honey production potential of the area, all the different segments of the honey chain should be addressed starting from input supply and improved extension services to the consumption; through providing skill development trainings, enhancing input supply chains and market channels and challenging the financial constraints.

[P.18.435] Mapping Apiculture Value Chain and Gender Roles in Southwest Nigeria

A. Balogun, F. Dada

Federal University of Technology - Department of Meteorology & Climate Science, Akure, NIGERIA

National consumption of honey is estimated to be about 380,000 tons annually, while National production is not up to 10% or 38,000 tons. This gap is almost 340,000 tons or at least $2 billion!

Establishing Nigeria's competitive advantage in honey and beeswax production to develop a substantial diversion from the current unsustainable oil based economy is an important goal of the Agricultural Promotion Policy, 2016 - 2020 (FMARD, 2016). Increasing productivity and incomes in the very large and small-scale commercial apiculture sector is part of the long term vision.

Nigeria has comparative advantage for beekeeping or apiculture. There exists a huge variety and abundance of melliferous plant species in Nigeria serving as forage for bees). Due to bimodal rains in southwestern Nigeria, honey can be harvested at least twice a year.

The apiculture sector in Nigeria, however, is far from realizing its potential for earning foreign exchange, as well as generating income for smallholder beekeepers and other actors in the value chain. Less than 10% of the honey and wax potential have been tapped, and the commercialization of other high value bee products such as pollen, propolis and bee venom is non-existent. Addressing these issues through a private sector driven value chain and market system approach will go a long way in realizing the full potential of apiculture in the country. However for this to be achieved information on the constraints, gaps and investment opportunities in the apiculture value chain need to be identified. This mapping study aims to provide this information by conducting surveys in selected local government areas of Ogun, Ondo, Ekiti, Osun and Oyo state in southwest Nigeria. The survey will cover demographics of the apiculture value chain actors and their relationships with each other, inputs, education, training and extension services, production systems, products and processing, packaging, finance, markets and marketing.

The results of this study will provide recommendations for business investments the apiculture industry in southwest Nigeria.

[P.18.436] Economic and financial integration of young people: challenges and prospects

P. Sefu Wa Sefu

Groupe Stigress Sa, Kinshasa, DEMOCRATIC REPUBLIC OF THE CONGO

Entrepreneurship and living together are the means of economic and financial inclusion of young people. The more young people create small and medium-sized businesses, the more the state becomes richer; Starting a small business is one way to participate in the economic life of your own country.

Africa needs competent, courageous and ambitious young people who are irreversibly involved in its integral development. It is not forbidden for Africa to be resourceful in the West and in the East for its development. But, ideally, a development that is rooted more easily if it depends on endogenous factors and actors.
Hence, the imperative need to support young people who forge the spirit of entrepreneurship and creativity. Motivation and support will allow them to go further in their initiatives.
Presumably, economic development does not happen overnight: it is an area that requires a spirit of perseverance in business and creativity. It intervenes much more as young people get up to fight unemployment and idleness.
However, to undertake, it is not enough simply to have a colossal amount as most young Africans think. A single step is enough for a click that allows a long trajectory.

[P.18.437] Geographic Indication of bracatinga honeydew honey: a tool to promote product competitiveness

D. Dortzbach, E. Vieira, E. Blainski, V.F. Vieira, K. Trabaquini, E.B. Da Silva, F.M. Zambonim
Epagri, Florianopolis, BRAZIL

Honey of bracatinga honeydew (Mimosa scabrella Bentham) is obtained from mealybugs (Tachardiella sp) that feed on the sap of the plant and excrete the excess of honeydew droplets that are collected by the bees. This honey has specific characteristics that differentiate it from the others, due to the strong characteristic flavor and odor, darker color when compared to floral honeys, higher electrical conductivity, lower monosaccharide content (glucose and fructose), a possible reason why it usually does not crystallize. Considering the specific characteristics of the honeydew honey produced in South of Brazil, along with tradition, culture and economic importance, the proposal of producers with the support of Epagri and Sebrae for the implementation of a geographical indication (GI) is presented, which is one of the valorization tools of products with territorial specificity and that can also contribute to the development of the affected territories, mainly in the social, cultural and environmental dimensions. The implementation of the GI would signal to the consumer that it is a genuine product, whose specificity is due to its origin, conferring a market differential. This is a necessity, since with globalization, consumers around the world are increasingly interested in foods with guaranteed local and regional identities, and these certifications are appropriate to ensure higher quality, safety and to address specific market niches. In this sense, the addition of the differentiation and specific market, the aggregation of value to the product, incorporating to the same, the history of the region, the culture, the know-how and the local identity is obtained through the GI, offering to the consumer a sign of its typicality and quality.

[P.18.438] Devolopment of Bee Products Processing Industry in China

B. Fang, R. Li
Institute of Apicutural Research, Beijing, CHINA

China's bee product processing industry is developing steadily, and the transformation and upgrading has achieved preliminary performance. Enterprises licensed to produce bee products in 2017 gathered in raw material producing and trading areas. Affected by the policy of poverty alleviation, the distribution of bee products processing industry shifted to poor areas with better ecological environment. Common edible bee products continue to be dominated by honey, and the unique position of propolis in health-care bee products is further consolidated, and the processing of bee products tends to natural maturity and original ecology. The scale of the enterprises is mostly small and medium-sized, and the net profits are generally not high. More than 80% of the bee products enterprises surveyed do e-commerce business and maintain a high growth rate. The number of professional beekeeping cooperatives in China has reached 13040, and the management system has been further standardized, which is a new force in bee product processing industry. There are also some problems in the steady development of China's bee product processing industry. In the future, efforts should be made to improve the modern management level of enterprises, increase investments in research and development, promote the transformation and upgrading of bee e-commerce and enhance the credibility of enterprises

[P.18.439] Presentation of the process of helping beekeepers in Slovenia with a consulting service

L. Senic
Slovenian Beekeeping Asociation, Lukovica, SLOVENIA

In Slovenia, beekeeping is given a great deal of importance. The authorities are aware of this, therefore they financially support beekeeping through various services that provide beekeepers with certain benefits. One of these services is the public beekeeping advisory service (PBAS), which operates throughout Slovenia. Established in 2008, it offers general on-field consulting to beekeepers, in addition to specialized consulting. Specialized consultations are carried out by 7 specialist consultants, whereas on-field consultations are performed by 19 field consultants.
The basic purpose of the Public beekeeping advisory service is to advise beekeepers in the field of beekeeping technology, food safety, economics of beekeeping and other tasks. The program follows of goals set out in the strategic documents, among which are the conservation of the indigenous population of the Carniolan bee, the preservation of sufficient population of bee families, the preservation of high quality and variety of bee products, the improvement of technology and the economy of beekeeping.

The Republic of Slovenia granted a concession to the Beekeeping Association of Slovenia for the implementation of PBAS in the field of apiculture. Specialist consultants and field consultants operate according to the program, which is adopted annually by the Government of the Republic of Slovenia. Specialist consultants have in-depth knowledge in the field of beekeeping, therefore they are training, advising and informing beekeepers and field consultants with internet articles and articles published in the professional journal Slovenski cebelar, brochures, lectures, workshops, seminars and technical video footage. Specialist consultants also perform internal control of honey, which includes the determination of physical-chemical, microscopic and sensory properties of honey. Field advisors are the right hand of specialist consultants, they offer on-field help to beekeepers directly by acting on the beekeeper's requests, especially by providing practical tips for beekeeping.
Specialist consultants and field consultants must meet certain conditions defined by level of education and experience. Specialist consultants receive further education themselves by studying professional publications and by participating in professional conferences home and abroad. They are also engaged in research work in the field of beekeeping, the conclusions of which are passed on to beekeepers and field consultants.

[P.18.440] The importance of cooperativism for geographical indication - bracatinga honeydew honey

D. Dortzbach, E. Vieira, E. Blainski, K. Trabaquini, V.F. Vieira
Epagri, Florianópolis, BRAZIL

The Santa Catarina beekeeping association, through the Federation of Associations of Beekeepers and Meliponicultores of Santa Catarina (FAASC), founded in 1979, is an example of successful integration, bringing together 55 affiliated associations in every state. This organization is the key element for a geographical indication (GI) because it is the same as the referral process of the National Institute of Intellectual Property (INPI) and that of IG, without the purpose of meeting the needs of the honeydew honey production chain of the Santa Catarina Plateau. To the quality of the best the effect of the compliance of goon and regulated in the first list of the compliance of the product and public regulated the product. The justification is the request for the merits of the same, if the notification of product, the local of original, and the quality and unique binding bound to environment, already already valued not outside. Allied to this, increased competition and the need for consumers, the need to seek ways of insertion and stay in the market increasingly competitive. Thus, an association has been a viable alternative in the attempt to obtain a list of producers in search of product valorization. Among the challenges of a GI are benefits for producers, as well as apiculture associations and honey beekeepers, as well as honeydew honey. There is currently an area for obtaining multiple companies and for success. Thus, it is essential that the management of a GI is managed by an entity that has the representation of companies throughout the State.

BEE BIOLOGY

09 SEPTEMBER 2019

POSTER SESSION 02 08:30-18:00
BEE BIOLOGY I POSTER AREA

[P.02.07] Genetic selection of Iranian honey bee (*Apis mellifera meda*) for honey production, swarming and defense behavior during sixteen generations

G. Tahmasbi [1], M.A. Kamali [2], R. Ebadi [3], M. Babaei [1], H. Baneh [1], A. Gharadaghi [4], H. Afrouzan [1], N. Tajabadi [1], E. Seyfi [1], P. Valizadeh [1], S. Parichehreh [1]
[1] *Honey Bee Department, Animal Science Research of Iran, Agricultural Research Education and Extension Organization, Karaj, IRAN,* [2] *Agriculture research and education organization, Tehran, IRAN,* [3] *Plant protection Dep. Agriculture college, Isfahan university of technology, Tehran, IRAN,* [4] *Deputy of Animal Affairs, Ministry of Jihad-Agriculture, Tehran, IRAN*

Due to the behavioral and biological characteristics of honey bees and their dependence on environmental factors, the most appropriate breeding strategy is to use native races in different regions of the world. Iranian honey bee (*Apis mellifera* meda) has lived in Iran millions of years before the humans. Therefore, the Iranian Honey Bee Breeding Program was selected as the main strategy of beekeeping industry of Iran. First, the survival of Iranian honey bee and three different populations of them were proved using morphological and biochemical characteristics. Breeding Program was started in Tehran, Markazi, Isfahan and Qazvin provinces in 1998. In this presented research honey production, swarming and defense behavior were evaluated in 5000 native populations of *A. mellifera* meda followed by selecting the superior 20 % and transferring them to the Animal Science Research Institute of Iran, located in Karaj (Alborz province). In order to stablish the next generation during the 16 generations of the program every year (except 2002, 2010, 2012 and 2014), the most superior evaluated 40 drone producer and 100 mother colonies were selected. This research demonstrated that the genetic and phenotypic trends of honey production, swarming and defensive behaviors in the honey bee colonies were desirable during the 16 generations. The mean results of the first three and the last three generations showed that the queen cell numbers were decreased from 1.59 to 0.08 and also the sting numbers were deceased from 13.96 to 1.057. In other words, the mentioned traits have improved more than 10 times during the 16 generations of the Iranian Honey Bee Breeding Program. Moreover, the comparisons revealed an increase of almost 3 times in honey production. The results demonstrated that the genetic and phenotypic trends of different traits in the honey bee colonies were desirable during the 16 generations. In this study performance of the bred queens and control queens were compared in private apiaries in 2006, 2007, 2009, 2013 and 2016. All these findings confirm the desirable improvement of honey bee colonies in the Iranian Honey Bee Breeding Program.

[P.02.08] Sperm traits of male solitary bees, *Osmia cornuta*

V. Strobl [1], L. Straub [1,2], S. Bruckner [1,3], M. Albrecht [4], J. Maitip [5,6], E. Kolari [1,2], P. Chantawannakul [6,7], G.R. Williams [1,2,3], P. Neumann [1,2]
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Reproductive strategies can select for reproductive traits of males. Therefore, variation in sperm quantity and viability between bee species can shed light on the mating system. However, measures of sperm traits are lacking for solitary bees. Here, we evaluated for the first time sperm quantity and viability in male *Osmia cornuta* at different times after emergence, and how they were affected by male body mass and environmental conditions (laboratory or semi-field arena). Overall, O. cornuta males produced on average 175'000 spermatozoa that were ~65% viable, which are both significantly lower compared to eusocial honeybees and *Bumblebee*s. Sperm viability after emergence was not significantly different compared to four day old males, supporting that mating can occur immediately. However, sperm counts were significantly higher in four day old individuals from the semi-field arena when compared to newly emerged males, possibly reflecting sperm maturation. Moreover, sperm quantity, but not viability, was positively correlated with male body mass four days after emergence. Even though individuals maintained in semi-field conditions exhibited a significantly greater loss of body mass, experimental arena had no significant effect on survival, sperm quality or total living sperm produced. This suggests that the proposed laboratory design provides an adequate approach to assess sperm traits in solitary bees. In conclusion, our data suggest a reduced investment in sperm quantity and quality by male O. cornuta, which could be adaptive in light of the life history of this solitary bee.

[P.02.09] Histological characterization of testicular development of *Apis mellifera* drones during sexual maturation

C. Klein, S. Derveau, I. Kozii, S. Wood, R. Koziy, I. Dvylyuk, E. Simko
Western College of Veterinary Medicine - Department of Pathology, Saskatoon, CANADA

Background: Current risk assessment procedures for the exposure of pollinators to pesticides are not adequately predicting the detrimental interactions reported in scientific literature. The "gold standard" for compound risk-assessment relies heavily on histopathology in

mammals, but it is not employed in honey bees.
Objective: To characterize the normal morphological changes of testes of honey bee drones during the sexual maturation period in order to establish a normal benchmark for future gonadotoxicity studies.
Methods: At emergence, drones were marked and subsequently sampled each day during the period of sexual maturation (2 weeks). The reproductive tract of each drone was dissected, photographed and analyzed macroscopically and microscopically.
Results: Drones undergo sexual maturation for 2 weeks following emergence. The mature sperm moves from the testes to the seminal vesicles via the vas deferens, while the testes undergo progressive involution and atrophy. The testicular changes during sexual maturation are characterized by several distinct processes: 1) Spermatogenesis continues after emergence for 3 days; 2) By day 8, the entire sperm content is moved from testes to vas deference and seminal vesicles; 3) Testicular parenchyma undergoes degeneration and apoptosis followed by complete parenchymal loss by day 11 – 15 (depending on environmental temperature); 4) The end-stage testes consist of collapsed supporting stroma containing brown pigment and tracheal network. The testicular involution was accelerated by approximately 4 days in drones examined during warmer months.
Our future studies will evaluate effects of insecticides on gonadotoxicity in drones exposed to neonicotinoids during the sexual maturation period.

[P.02.10] Uncovering the new world of non-coding RNAs in two widespread fungal pathogens of honeybee

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Ascosphaera apis and Nosema ceranae, two widespread fungal pathogens of honeybee, are main threats for bee health and cause heavy losses for apiculture. Currently, non-coding RNAs (ncRNAs) including long non-coding RNA (lncRNA) and circular RNA (circRNA) are hotspots attracting increasing attention from researchers. However, compared with animals and plants, knowledge of ncRNAs in fungi is extremely limited. Here, mycelia and spores of A. apis and purified spores of N. ceranae were separately sequenced utilizing next-generation technology followed by systematic analysis with bioinformatic approaches. 379 lncRNAs were first identified in A. apis, including antisense lncRNAs, long intergenic non-coding RNAs (lincRNAs), intronic lncRNAs and sense lncRNAs; while 83 novel lncRNAs in N. ceranae, including lncRNAs, lincRNAs, and sense lncRNAs. Notably, these lncRNAs share similar characteristics with those identified in mammals and plants, such as shorter length and fewer exon number and transcript isoforms than protein-coding genes. Additionally, several predicted lncRNAs were randomly selected for RT-PCR assay, and the results validated their true existence. Interestingly, expression levels of 11 lncRNA in A. apis were found to be lower than those of adjacent protein-coding genes. Meanwhile, 551 novel circRNAs were first identified from A. apis, including 134 exonic circRNAs and 416 intergenic circRNAs; while 2014 from N. ceranae, including 174 exonic circRNAs and 30 intergenic circRNAs. Most of the circRNAs were 200–600 bp in length, which were different from animal and plant circRNAs. In addition, the expression of several predicted circRNAs was confirmed by RT-PCR. Moreover, circRNA-miRNA regulation networks indicated both A. apis circRNAs and N. ceranae circRNAs could regulate gene expression by absorbing miRNAs as competitive endogenous RNAs (ceRNAs). Functional analyses of the miRNA target genes demonstrated these fungal circRNAs were likely to play pivotal roles in growth, development, metabolism, environmental response and gene expression. Taken together, this is the first comprehensive study of ncRNAs in A. apis and N. ceranae, opening the door for the mysterious world of fungal ncRNAs. Our findings offer invaluable data resource for further study on fungal lncRNA and circRNA, and provide novel insights into understanding the basic biology of A. apis and N. ceranae.

[P.02.11] New protocols to study nectar foraging of honey bees

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Based on new technological developments, we propose new automatized protocols whose purpose is to control the exposure to the nectar resource and to measure in detail the nectar foraging behavior of honey bees.
Free flying - individually marked- honey bees are trained to come to visit a computer controlled artificial flower that gives access to small and variable amounts of syrup. First, we show that the nectar flow entering the colony is constant over time and decreases when nectar resources decrease over time. Then, we show that pesticides may affect this nectar flow. Finally, we show that toxicity of nectar may increase when resources become more scarse in the environment.
As nectar flow entering the colony is a dependent variable highly linked to colony fitness and beekeeper economic health, we suggest to assess this flow when testing the non lethal and chronic or subchronic effects of pesticides.
In conclusion, we suggest some new directions to improve our device and protocols to be able to give a more complete possible description of pesticide non lethal effects.

[P.02.12] *Apis mellifera capensis* together with the *scutellata* causes the africanisation

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In Brazil we have a gentle, africanized, dark brown bee, which is predominant in colder and humid regions. In hotter and dryer regions, the

africanized yellow bee is predominant, showing an extraordinary defensivity.
With increasing commercial beekeeping in the last 20 years , these two types of bees have cross-bred. The result was negative: more defensivity , swarming at any time, frequently self-requeening , losing queens and laying worker bees.
We would like to know what is the difference between the dark brown bee and the yellow bee? It is a fact that, in 1956, Dr.Kerr brought 15 *Apis mellifera* capensis to the University of Rio de Janeiro, where Prof.Manoel Barros inserted them into 15 European beehives. Three months later these beehives were dead! His first conclusion was, that these brown bees were not adapted to the climate. But two years later he discovered that, around the University region only dark brown bees were present and the European bees had completely disappeared. This was the effect of Teletokia. The capensis worker bees were accepted as queens! Better: Pseudo Queens. They have the ability to lay fertilized eggs and maintain viable clonal lineage for more than twenty years! Plenty of time to exterminate the established European bee.
At the same time Dr.Kerr was working in Rio Claro, Sao Paulo, with the scutella bee. In the beginning it was a success, because the scutella virgine queens were fertilized by European drones. The result was a high production of honey. Prof.Kerr and other beekeepers started to breed scutellata queens. But the problem was that the European drones disappeared. They were simply made extinct by the capensis.
Now the conflict between capensis and scutellata was obvious. This began in the 1970's and became a big problem. Mainly for the commercial beekeepers. This proves, that the capensis and the scutellata caused the africanisation. For Brazil, this is neither a mystery of the past, nor a doubt of the present and the future. Attention! This bee is frost resistant and can be confused with the European bee by producers of queens in South America!

[P.02.13] Genetic differentiation in honey bees, a new tool for subspecies conservation and breeding

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Europe is home to a wide diversity of honey bees, differentiated into 13 subspecies. Across their natural range honey bees have adapted to the variable environmental conditions: climate, flowering patterns and disease strains. Many european beekeepers are actively involved in conservation programs to protect their local subspecies. Due to the movement of honey bees for honey production and pollination, and the trade of queens, hybridisation is likely to occur. It is essential to be able to discriminate between pure subspecies and their hybrids, for the sake of conserving our diversity, but also to secure their adapted traits.
SmartBees, a project funded by the European Commission, had the dual goal of protecting the European diversity of honey bee subspecies, and to assist the beekeepers in breeding varroa tolerant bees for all the subspecies. To achieve this purpose we had to develop new tools for subspecies differentiation and varroa tolerance based on next generation sequencing.
To this end, more than two thousand samples of all subspecies have been collected across Europe, representing the total *Apis mellifera* genetic diversity on the continent. Next generation sequencing was performed on twenty-two pools each of more than 90 worker bees, representing different populations of honey bees, from Portugal and Ireland in the west, to Russia and Armenia in the east. Based on the pool sequence data a new molecular tool, a so-called SNP (single-nucleotide polymorphism) chip was designed, consisting of highly informative markers for ancestry. The SmartBees SNP chip contains several thousand SNPs to describe the genetic diversity of individual samples, related to subspecies diversity and traits related to varroa tolerance. The assignment power for subspecies has been tested with more than 6000 samples and allows for the detection of pure subspecies and their hybrids. We have demonstrated that pure subspecies are correctly predicted in 98% of the cases.
Determination of the subspecies' heritage of individual bees is highly relevant in order to achieve conservation goals and breeding progress.

[P.02.14] Expression pattern of Cytochrome P450 343a2, in the Honey Bee, *Apis mellifera*

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Cytochrome P450s (CYP450s) in honey bee, *Apis mellifera*, not only play a crucial role in detoxification against various pesticides and phytotoxins but also involve in insect hormone biosynthesis. In our imidacloprid-affected transcriptomic library shown that one gene fragment of CYP450 encoded GB43693 was induced significantly. The corresponding cDNA was cloned and found to have 95% identity with the CYP450 343a1, named AmCYP343a2. AmCYP343a2 was classified into the CYP2 family whose members might be involved in hormone biosynthesis and developmental processes of organ. Quantitative RT-PCR measurement of the AmCYP343a2 expression in the heads revealed that the gene expression reaches peak during late pupa stage and can be regulated by 20-hydroxyecdysone and the juvenile hormone analog, pyriproxyfen. We further analyse the expression of AmCYP343A2 in the honey bee head reveals that the mouthparts and cuticle, have the highest expression comparing to mushroom body and hypopharyngeal gland. Taken together, it was speculated that AmCYP343A2, the gene expression induced by imidacloprid treatment, may play a specific role in the development of insect epidermis during pupa stage.

[P.02.15] 100 years of the journal Bee World

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The journal Bee World was founded in 1919 by Dr. Ahmed Zaki Abushâdy, an Egyptian living in England, and the Apis Club. Publication was taken over by the International Bee Research Association (IBRA) in 1952. With the exception of a hiatus between 2006 and 2009, it appeared uninterrupted, and is published in its 96th volume in 2019. So far, more than 6,500 articles have been published in the journal. In 2015, Taylor and Francis took over the publication of Bee World, which is published as four issues each year. Among others, Original Articles, Review Articles, Art & Culture, Book Reviews, Obituaries and Conference Abstracts are published. Bee World is a printed journal, but all articles, including those from all editions going back to the very first issue, are also available online (see www.tandfonline.com/toc/tbee20/current). Bee World is IBRA's popular journal, available free to IBRA members, and acts as a bridge between today's beekeepers and bee scientists, encouraging two way discussions. There are no publication fees. In 2018, an editorial board was established, one that seeks to develop the journal further. Special focus will be placed on the 100th anniversary of Bee World in all four issues published in 2019. The poster at Apimondia will display the most influential articles published in Bee World, with respect to the scientific impact (number of citations) and societal impact (number of article reads). The editor and editorial board invite you to submit your article or to discuss possible contributions to Bee World.

[P.02.16] Antioxidant and antityrosinase activities of Thai monofloral bee pollen

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Bee pollen of *Apis mellifera* was collected from tea flower (Camellia sinensis (L.) Kuntze) and mimosa flower (Mimosa pigra L.) in Chiang Mai province and sunflower (Helianthus annuus L.) from Lopburi province, Thailand. All samples were extracted by methanol and were, next, partitioned by hexane, dichloromethane, and methanol in order to isolate compounds depending on their polarities. The obtained partitioned extracts were tested for the antioxidant and antityrosinase activities. For the antioxidant activity by DPPH assay, dichloromethane partitioned extract of mimosa flower bee pollen (DCMMBP) provided the highest antioxidant activity at EC50 value of 192.07 µg/mL. Then, it was further purified by silica gel 60 column chromatography and size exclusion chromatography. All fractions were tested for the antioxidant activity and analysed for a chemical structure by nuclear magnetic resonance (NMR). The most active mixture had the EC50 value of 121.29 µg/mL. Additionally, a pure compound was found to be naringenin, belonging to flavonoid group. For the tyrosinase inhibitory activity, it was tested by a biochemical reaction of an extract on mushroom tyrosinase. It showed that dichloromethane partitioned extract of sunflower bee pollen provided the highest mushroom tyrosinase inhibitory activity at IC50 value of 159.39 µg/mL. After further purified by silica gel column chromatography and HPLC, all fractions were tested for antityrosinase activity and analysed a chemical structure of active fractions by NMR. The active mixtures were predicted to be spermidine derivatives which provided the highest tyrosinase inhibitory activity at IC50 value of 6.65 µg/mL. It could be concluded that the antioxidant scavenging and antityrosinase agents were rich in mimosa flower and sunflower bee pollen, respectively.

[P.02.17] In vitro anti-tyrosinase activity of pure compounds from bee foraging plants

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Tyrosinase is the key enzyme in melanogenesis leading to skin darkening. Thus, many skin whitening products contain tyrosinase inhibitors or extracts. Recently, natural extracts are popular alternatives because they are safe or have less side effects. Here, fifteen compounds from 6 different plants including pinostrobin, pinocembrin, cardamomin and alpinetin from Boesenbergia rotunda, piperine from Piper nigrum, rosmarinic acid from Orthosiphon aristatus, methyl lansioside C, lansioside C and lansioside B from Lansium parasiticum, sesamin, sesamolin and samin from Sesamum indicum, malabaricone A, horsfieldone A and maingayone D from Horsfieldia motley were used. To determine the effects of these compounds on melanogenesis, tyrosinase activity was evaluated in vitro. Kojic acid was used as positive control. It revealed that piperine, horsfieldone A, and maingayone D had high tyrosinase inhibition with IC50 values of 0.526, 0.294 and 0.038 mM, respectively. Compared with kojic acid (IC50 = 0.059 mM), maingayone D showed the highest inhibitory effect against mushroom tyrosinase, while piperine and horsfieldone A were less potent. In cytotoxicity test, maingayone D exhibited cytotoxicity against B16F10 melanoma cells at concentration of 1.95 µg/ml (or 0.004 mM) with cell viability decreasing to approximately 80% of control after

72 h treatment. Horsfieldone A exhibited cytotoxicity against B16F10 melanoma cells at concentration of 3.9 µg/ml (or 0.011 mM) with cell viability decreasing to approximately 80% of control. Piperine showed toxicity at a concentration of 12.5 µg/ml (or 0.044 mM) with a cell viability of 80%, whereas kojic acid showed toxicity at a concentration of 500 µg/ml (or 3.52 mM) with a cell viability of 80%. Furthermore, IC50 values of piperine, horsfieldone A, maingayone D, and kojic acid were 0.079, 0.021, 0.019 and 10.765 mM, respectively. From this study, it suggests that piperine, horsfieldone A, and maingayone D are potential for tyrosinase inhibitors.

[P.02.18] Introduction to histologia of *Apis mellífera*

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The species *Apis mellifera* is one of the knownest species among the bees with sting, no native of Brazilian lands, being introduced in the southeast and south area of Brazil by Jesuits and Germans in the period of 1840. in this time the beekeeping was a differentiated activity, tends as initial objective the honey production to supply the farmer’s own need, with few handling techniques, being cultivated even in the back yard of the houses. Due to low aggressiveness of the European bees, his handling was placid and unperturbed, however, in his phase production he/she came low for the fact of they be accustomed with reduced temperatures, different from the Brazilian tropical climate. Starting from that she began researches more deepened by specialists of the area that Africa was destined in search of species more adapted, in the intention of comparing the productivity of the African bees with the Europeans, their habits, susceptibility of pathologies, appropriate climate and other information. Descending of the African bees brought by the researchers in this period to Brazil spread little by little in the Brazilian territory, where they crossed with the Europeans and we have the bees africanizadas today. it mixes of both. Being the most resistant africanizadas, more producing of honey and more aggressive.The body of the *Apis mellifera* is divided in three parts, head, thorax and abdomen, characteristics of the insects. Some works elucidate their organs and systems, as well as the glands hipofaríngeas, involved with the honey production, structures that you/they shelter the pollen collection and other morphologic characteristics, however, certain physiologic and histological characteristics of this insect, are still little studied. The present work has as objective the histological study of the systems involved with the honey production, própolis and other.

[P.02.19] Back to the Future of Swiss honey bees: Comparing the genetic diversity of the past and present, to inform the future

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Honey bees are particularly sensitive to inbreeding depression leading to reduced viability, while high intra-colony diversity increases productivity, disease resistance and survivorship.
Moreover, it has been shown that the genetic origin is important with locally adapted honey bees having higher longevity, docility and productivity. Given the multitude of pressures facing honey bees today and the need for future adaptability, it is crucial to preserve the diverse genetic resource that locally adapted honey bees provide. However, in a world where honey bees are largely managed what, precisely, defines locally adapted honey bees? What characterizes their pristine genetic identity and diversity?
Advances in sequencing technology enable us to sequence museum specimen to get new insights into the genomic past. Using Swiss *Apis mellifera mellifera* as a case study, our research provides important insights into the genetic diversity of native honey bees prior to the industrial-scale introductions and translocations of non-native stocks during the 20th century, the onset of intensive commercial breeding, and the decline of wild honey bees following the arrival of Varroa destructor. To this end, we have extracted DNA from hind legs of A. m. mellifera specimen from the Natural History Museum in Bern, Switzerland, and have successfully sequenced the whole-genomes of 21 samples dated between 1879 and 1959. This is the first study to examine whole-genome sequences of historic and modern honey bees from their native range.

[P.02.20] Genetic characterization of commercial honey bee, *Apis mellifera*, populations in Thailand by using microsatellite and mitochondrial DNA markers

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Process of domestication is associated with a profound reduction in genetic diversity. The honey bee, *Apis mellifera*, has been managed by beekeepers for centuries for both production (honey and wax) and economic crop pollination. Here we used both microsatellite markers and sequence data from the mitochondrial COI gene to evaluate the temporal genetic variation of managed *A. mellifera* sampling from 8 intensive stock area of commercial beekeeping in Thailand. Microsatellite analysis revealed the moderate low average genetic diversity (expected hetrozygosity, He). Additionally, most of observed heterozygosity values were lower than expected heterozygosity under Hardy–Weinberg equilibrium. Mitochondrial determination detected two evolutionary lineages (C and O lineages) with low genetic distance ($0 < Fst < 0.034$) but high migration rate ($10 < Nm$) between pairs of commercial apiaries and stay the same result comparing with the assessment study in Thai commercial *A. mellifera* population from the last decade. We also detected no evidence of recent changes in effective population size (Tajima's D = –0.25, P > 0.05; Fu's Fs = –3.75, P > 0.05), indicating no recent major change in propagative population. Our results demonstrated that Thai's manage *A. mellifera* may heading to reduce genetic diversity. The admixture management might be considered to promote genetic diversity of the population is suggested.

[P.02.21] The Welsh Dark bee (*Apis mellifera mellifera*) is not extinct

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Due to past and present imports of *Apis mellifera ligustica* (Italian bees), *Apis mellifera* carnica (Carniolan bees) and the English Buckfast bee (a hybrid strain) across its entire natural range, *Apis mellifera mellifera* is now threatened with extinction by genetic pollution through hybridization. Whilst the status of remnant A. m. mellifera populations is well documented on the European mainland, few studies have been undertaken to identify surviving populations on the British Isles. A few A. m. mellifera stocks are thought to persist in Scotland and the southwest of England and recently, Ireland might appear to contain multiple non-hybridized A. m. mellifera populations. 163 young worker bees, representing 121 colonies from across Wales, were genetically screened in an attempt to identify remnant A. m. mellifera stocks, as part of a conservation breeding program. Recent studies have demonstrated that honey bees of local origin have significantly higher survival chances than honey bees of non-local origin due to their adaptation to their local environment, suggesting that conservation of locally adapted honey bees is a logistical and practical possibility to develop sustainable apiculture. Within this survey, we made use of a custom-tailored SNP genotype assay to estimate the extent of C lineage introgression in the ncDNA as well as sequencing of the tRNAleu-cox2 intergenic region of the mtDNA to check for the ancestry of the tested colonies.

[P.02.22] Quantifying the relationship between genome-wide colony-level genetic diversity and honey bee health and productivity

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A number of studies have investigated the relationship between social insect colony-level genetic diversity and measures of colony health and productivity. However, such studies have been conducted with small sample sizes, limited numbers of genetic markers and extreme experimental designs, such as comparing colonies headed by singly-inseminated queens to those headed by multiply-inseminated queens. Using 918 colonies situated across Canada, we investigate the relationship between genome-wide colony diversity and various phenotypic measurements, such as fall cluster size, fall weight, viral load and winter survival. Results of this study will help guide researchers and beekeepers by quantifying the importance of genetic diversity with respect to honey bee health and productivity.

[P.02.23] Efficacy of DNA barcoding for identification of female castes of Bombus ignitus and B. ardens using cytochrome oxidase I and their genetic variations

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Bumble bees are important pollinators of many wild flowers and economic crops. Among Korean bumble bees, female castes of *Bombus ignitus* and B. ardens are morphologically similar sharing black head and thorax and abdominal tergites 1-3 while the abdominal tergites 4-6 of both species are yellow. Drones are easily distinguishable, but foraging females are not. To test the efficacy of DNA barcoding method for identification of female castes of these species, 39 females of both species were collected in South Korea and sequenced using a 653 bp fragment of the cytochrome oxidase subunit 1 (CO1) region of mitochondrial DNA. 33 samples of B. ignitus were obtained from three commercial populations in Yecheon, Sangju (South Korea) and Japan, to compare the sequence variations and haplotype diversity. The result showed that DNA barcoding is an efficient method to differentiate B. ignites and B. ardens. In addition, Yecheon population with three haplotypes had the highest sequence variation in the studied populations of B. ignites. Sequence variations of 653 bp of COI gene of B. ardens collected in this study and 284 previously recorded sequences of Korean populations showed 13 haplotypes with pairwise nucleotide variation between 0.16 - 0.32 % while sequences variation in 448 bp of the same gene of B. ignitus collected in this study and 401 previously recorded sequences of Korean, Japanese, Chinese and Belgium populations showed 18 haplotypes with 0.21- 2.01% pairwise nucleotide variation.

[P.02.24] Genomic tools for tracking invasive Africanized honey bees

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The honey bee, *Apis mellifera*, is an ecologically and economically important species contributing to pollination services worldwide.

Consequently, it is essential that potential threats to honey bee populations are identified and mitigated to prevent losses to the beekeeping industry. Africanized honey bees (AHB) are a hybrid population composed of European and African ancestry and are considered undesirable for beekeeping due to their aggressive defensive behaviour. Given the large-scale trade and movement of honey bees, there is a concern that AHBs will spread from South America and the southern United States to the rest of North America, Australia, New Zealand, and Hawaii. Developing an accurate and cost effective assay to detect AHB is an important first step towards restricting the accidental importation of AHBs. Here, we used an extensive population genomic dataset composed of individuals from all known evolutionary lineages to assess the genomic composition of *Apis mellifera* populations, and patterns of genetic admixture in North and South American commercial honey bee colonies. Our genomic dataset includes over 150 newly sequenced individuals from at least 14 subspecies encompassing the known distribution of *Apis mellifera*'s native range. We used this data set to develop a SNP assay that shows high accuracy in assigning bees of unknown genetics as either African or non-African. Our SNP assay has been validated on over 2000 individuals from commercial colonies located in Canada, Texas, and Australia, as well as feral colonies from Texas, and Brazil.

[P.02.25] Immune-related gene expression in honeybees (*Apis mellifera*) exposed to alpha-pinene and diallyl disulfide

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Synthetic acaricides for Varroa control may show the negative effects on honeybee physiology. Recently, Varroa resisted to synthetic. As a consequence, botanicals are being seriously considered as varrocide. However, some botanical have shown negative effects on hygienic behavior. To investigate the effects of new botanical varroacide, here, the effects of alpha-pinene and diallyl disulfide as plant secondary metabolites on honeybee immune gene expression were evaluated. Honeybee workers (1–3 days old) were collected and transferred to the laboratory. Before commencing, bee workers were fed with 20% sucrose solution. Bees were kept at 28 ± 2°C and 58 ± 5% R.H. Subsequently, the bees were exposed to LC10 and LC25 of alpha-pinene and diallyl disulfide using a feeding method. Prior to exposure, sublethal concentrations were individually dissolved in 20% sucrose solution and poured into cotton. This sucrose source was put in a Falcon™ tube, and then bees (n = 50) were released. For each treatment, 150 bees (450 bees, 3 replications) were kept. In control, only sucrose solution was used. On the 5, 10 and 30th day after treatments with LC10 and LC25 of alpha-pinene and diallyl disulfide, the surviving bees were collected and immunogenes levels (relish, abaecin, Dscam, BASK) were determined. The results showed that Dscam and BASK were significantly down-regulated while other genes were not significantly expressed. The effect of LC10 and LC25 and days after treatments were not influenced on gene expression. While we detected the down-regulation of Dscam, and BASK, the bee immune system seems unaffected by alpha-pinene and diallyl disulfide.

[P.02.26] Genetic Stock Identification of *Apis mellifera* introduced in China Based on Forensic Markers through STRs Linkage Group

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Animal microsatellite referred commonly as short tandem repeats (STRs) is only resource of DNA markers for establishing meiotic map on the chromosomes in *Apis mellifera*. The density of the third-generation map (AmeMap3) is characterized with 2008 STRs loci, as contributed greatly to the ultimate assembly of honeybee genome (version 4.0). To genotype stock of *Apis mellifera* introduced in China for more than one hundred years through forensic markers of STRs linkage group, we validated the availability of STRs loci involved in the research on *Apis mellifera* using microfluidic DNA chip in high through-put screen. First of all, we assessed PCR amplicons of 96 STRs loci applied widely in forensic studies of *Apis mellifera* (a). Furthermore, we selected a group of 90 STRs loci from 2008 loci of AmeMap3 in the study on genomic assembly (b). Finally, we carried out a genotyping assay for genetic stock of *Apis mellifera* at the level of 16 chromosome dependent STRs loci (c). It is shown from separate pattern of DNA in microfluid matrix that (a) the available PCR amplicons of STRs loci is significantly different from unavailable PCR amplicons of STRs loci; (b) 90 STRs loci were selected from PCR amplicons of 2008 STRs loci; and (c) a group of 16 STRs loci located respectively along different chromosomes were used to act as informative forensic markers for genetic stock identification among 100 honeybee samples. It is suggested from the experimental data that polymorphic alleles of STRs loci on the latest version of AmeMap3 is used to provide forensic markers for characterizing its genetic stock identification.

[P.02.27] Distribution of *Apis mellifera* mitochondrial DNA lineages in two Italian regions estimated using honey DNA analysis

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Different *Apis mellifera* subspecies are well adapted to a wide spectrum of ecosystems. *A. mellifera* subspecies have been grouped into four main evolutionary lineages characterized at the mitochondrial DNA (mtDNA) level: the West and North European M lineage (including: A. m. mellifera); the C branch of the East and Central Europe and North-Mediterranean region (including A. m. ligustica); the Oriental lineage (O); and the African lineage (A), which groups the African subspecies. Conservation of this biodiversity is threated by beekeeping activities that have mainly favored the widespread dispersion of different subspecies and by the commercial distribution of hybrid queens. Thus, it is recognized that regional efforts to preserve local honey bee genetic resources are important in maintaining *A. mellifera* biodiversity. In this study, we evaluated the dispersion of different *A. mellifera* mtDNA lineages in two Italian regions: Emilia Romagna (in the North of Italy) which has prepared a new law to preserve A. m. ligustica; and Sardinia, which is an isolated Mediterranean island. To this aim we used a novel approach that took advantage from the possibility to define the entomological origin of the honey by analyzing the environmental DNA that can be recovered from this food matrix. A total of 100 and 65 honey samples were collected from Emilia Romagna and Sardinian beekeepers, respectively. Extracted DNA was analysed using a PCR based method that we developed from an informative mtDNA region that can distinguish the A, M and C lineages. The C mitotype was observed in 100% (Emilia Romagna) and 90% (Sardinia) of the analysed samples. The M and A mitotypes were identified in 17% and 10% (Emilia Romagna) and 10% and 12% (Sardinia) of the honey samples, respectively. In Emilia Romagna and Sardinia, 83% and 80% of the samples had only the C mitotype whereas the remaining samples showed different combinations of two or three mitotype lineages or only the A mitotype. This study provided a first dispersion map of the different *A. mellifera* lineages in these two regions. These results could be useful to design regionals program for the conservation of the autochthonous honey bee subspecies.

[P.02.28] Negative Selection in Social Insects

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Eusociality, characterized in part by cooperative brood care, and reproductive division of labour, evolved independently several times in insects. The evolution of eusociality has been hypothesized to lead to differences in the extent of both positive and negative selection. While population genomics studies of eusocial insects have so far focused on positive selection, there has been no study of the extent of negative selection in social insects, and its relationship to the evolution of caste- biased genes. To address this knowledge gap, my research will estimate the extent of negative selection in honey bees, bumble bees, and wasps, through analysis of published population genomic datasets. My study will compare the relationship between the strength of negative selection and caste-specific patterns of gene expression, and examine if the strength of negative selection correlates with the level of social complexity in this species triad.

[P.02.29] Discrimination of honeybee populations from Cyprus based on geometric morphometrics analysis of wing shape

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Cyprian honeybee (*Apis mellifera* cypria) is a distinct subspecies of the European honeybee. Previous studies, using mainly molecular markers, have shown that the honeybee population of Cyprus is relatively homogenous, although the introduction of queens or colonies from central Europe and Turkey has had some impact.
In the present study, honeybees collected from 15 different areas in Cyprus were analyzed by geometric morphometrics, using the coordinates of 19 indicators on the vein intersections of the right forewing. The coordinates of the landmarks were recorded and the two-dimensional x, y Cartesian coordinates were digitized using the tpsDig program. The landmark coordinates obtained from tpsDig were used in the MORPHOJ package.
A population from Greece (Lemnos Island) was used as an out-group for comparison. A total of 2400 honeybees (15 honeybees per colony from 160 colonies) were used. Procrustes analysis (PA), principal component analysis (PCA), and canonical variate analysis (CVA) were used to indicate the population variability among the studied samples. According to the PCA and CVA of pooled data from each locality, 12 of the 15 populations in Cyprus presented high similarity, while three (located on the south east of the island) clearly differentiated. All Cypriot populations differed from the control population of Lemnos Island. The most distinct of the three different populations was in an apiary for queen breeding where many queens from Europe, Asia and Africa have been reared. The rest two apiaries which were differentiated from the main cluster of Cyprian honeybees, had most of their colonies headed with queens introduced from the queen breeding apiary.
These results indicate the need for protection of the local honeybee, *Apis mellifera* cypria, against uncontrolled introduction and rearing of introduced honeybee queens in Cyprus.

[P.02.30] Where did the honey bee come from?

K. Dogantzis, T. Tiwari, I. Conflitti, A. Zayed
York University, Toronto, CANADA

The honey bee, *Apis mellifera*, is natively distributed through Africa, Europe, and parts of Asia. Consequently, the species occupies ecologically diverse areas, which has resulted in the diversification of many morphologically and geographically distinct subspecies. Although it is commonly agreed that the subspecies can be divided into at least five genetically distinct evolutionary lineages, there remains considerable contention regarding the species ancestral origin and subsequent expansion into its native range. Addressing this question and resolving current disagreements is important for understanding how the honey bee genome diverged to facilitate local adaptation across its distribution. Here, we used an extensive population genomic dataset, which consists of over 200 individual genomes from at

least 14 subspecies with an emphasis on newly sequenced individuals from Asia and Africa; including A. m. lamarckii and A. m. unicolor. Our study aims to disentangle the out-of-Africa and out-of-Asia debate, in addition to estimating the genes and pathways under positive selection that may be responsible for the honey bee's adaptation across distinctive ecological space.

[P.02.31] The Morpho Genotypes of the native honeybees in the Republic of Benin

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The honeybees are key pollinators which contribute in improving livelihoods in many communities. The bee population diversity determines its resilience and adaptation to biotic and non biotic factors of any region. Such biodiversity is assessed through Morphometric characteristics and genetic markers. In the Republic of Benin, honeybees samples from 94 established apiaries distributed in the entire country were morphometricaly characterized using 7 parameters and the COI-COII regions of mitochondrial DNA of each bee were sequenced. The morphometric data analyses indicated that the honeybees population of the Republic of Benin may be divided into three distinct morpho-ecotypes adapted to the different ecological conditions of the country. These are the Benino-dry-tropical-ecotype in the north, the Benino-Sudanian-ecotype in the central part and the Benino-Sudano-Guinean-ecotype in the south. The DNA COI-COII regions sequence analyses confirmed that the honey bee population of the country belongs to 3 different mitotypes corresponding to *Apis mellifera adansonii* (66%) *A. mellifera scutellata* (30%) and *A. mellifera iberiensis* (7%). But these genotypes do not correspond with the determined morpho-ecotypes. All found haplotypes miss the P0 segment. But their Q region is duplicated or triplicated and the Phylogenetic analyses clustered them together in the A evolutionary lineage. Morphometric and genetic analysis of the Benin native honey bees indicated that each of the different mitotypes were able to adapt to the different ecological conditions in the country by morphometric adjustments.

[P.02.32] Dissecting the genetic underpinnings of pathogen loads in the honey bee (*Apis mellifera*)

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The honey bee, *Apis mellifera* is a model organism for sociogenomics and is one of the most important managed pollinators. As such, recent threats to honey bee health are particularly alarming. The social honey bees live in highly crowded nests providing favorable conditions for the spread of infectious diseases. But honey bees have several social and individuals mechanisms for protecting themselves against disease. The BeeOMICS consortium has sequenced the genomes of approximately 1,000 honey bee colonies in Canada, which were evaluated for a number of traits, including the abundance of several pathogens within each colony. I plan to carry out genome-wide association studies (GWAS) on colony pathogen loads to gain a deeper insight into the genetics of immunity in honey bees. This research will set the groundwork for breeding disease resistant honey bees using marker assisted selection.

[P.02.33] Genetic diversity analysis of *Apis dorsata* honey bee species populations of districts nankana and narowal, Pakistan, by using rapd marker technique

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The study was planned to evaluate the genetic variations in genotype of two populations of giant honey bees "*Apis dorsata*" from two districts i.e. Nankana and Narowal from Punjab using RAPD-PCR technique. About 30 worker bees from each population were selected as samples and preserved in 50% alcohol till the completion of analysis. DNA extraction was performed by Phenol-Chloroform-Isoamyl method followed by observation on 1.0 % Agarose. DNA was amplified using ten random decamer RAPD primers. The amplification results were reproduced further using A-10, A-18, Moh-13 and OPA-07 against 1 kb reference ladder. The PCR products were analysed for their banding pattern based on the molecular weight. Five to eight bands were obtained with a range of 200 to 3000 bp size. Few polymorphic bands were found which showed that low level of genetic variability existed both within and between populations of Nankana and Narowal districts, respectively. Nei's analysis using POPGENE showed high level of homozygosity showing their origination from the same ancestor. However, Heterozygosity values showed that low level of genetic variations occurred in both Nankana and Narowal districts. It is, thus, concluded that there is variability in genetic profile among the populations of honey bees from two areas which could be due to human interference, climatic or nutritional differences.

[P.02.34] Molecular identification of bees in breeding and commercial apiaries in Greece

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Adult samples from different subspecies of *Apis mellifera* from bee breeders and commercial beekeepers were collected from many areas in Greece and they were subjected to COI mitochondrial gene segment sequencing analysis for identification purposes. The main objective is to have a clear view of the genetic origin of the bees used for breeding in Greece.
The results of that analysis may be used to possible pinpoint the geographical origin of honey bees according Ruttner (1988) and to indicate the known regions of the subspecies of *Apis Mellifera* in Greece. Based on the results there is also the possibility that a new geographical distribution of the managed honey bees used for bee breeding in Greece, to be mapped. The data obtained will contribute to the knowledge of the honey bee market in Greece as well.

[P.02.35] Designing a national Honeybee Breeding Objective: Dealing with diverse Beekeeper Requirements

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In a geographically and economically diverse environment such as New Zealand, the economic requirements of beekeeping operations differ greatly between locations. With the rise of Manuka honey as a high-value product in the international market, the corresponding beekeeping industry has undergone substantial changes throughout the past decade, with a tendency towards the establishment of large-scale corporate beekeeping operations that run hives in several parts of the country. Simultaneously, dedicated queen breeders have established themselves that will produce high numbers of queens (up to ~50,000), often from a small number of breeder queens. However, reports from the industry are indicating that this system is failing to deliver high quality stocks to some regions, potentially because the performance of these selected queens has only been evaluated in a specific environment. In the current situation, both queen breeders and commercial beekeeping operations buying in breeder queens are ill-equipped to supply or purchase queens that are well-suited to their conditions and management styles. These circumstances represent a prime opportunity for the implementation of a breeding objective that seeks to express the ideal individual in economic terms. Breeding objectives are in use across a wide range of agricultural species, with transformative results for the farming sector. We have developed a preliminary breeding objective for the New Zealand beekeeping industry, which will be used to derive tailored selection indices for beekeeping operations targeting high-value honey crops, low-value honey crops and honeydew. Since selection tends to be limited to ad hoc within-yard selection, personal preference plays a big role in queen breeding, but the economics of this are not well understood. In addition to the breeding objective built around available economic data on commercial beekeeping and the value of hive products, we used a decision-making and conjoint analysis software (1000minds) to conduct a survey of New Zealand beekeepers to understand personal preferences in queen and their underlying reasoning and integrate these in the breeding objective. This approach has been used in other livestock species with good results, and we hope that it will help close the gap between dedicated queen breeders and commercial operators.

[P.02.36] How Bee Breeders change the genetics of their stocks

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Breeders work hard to select the best bees for their customers, and many beekeepers rely on just one or a few breeders whose stock they think is best for their conditions. Just how much impact do breeders have on the genetics of the stocks they select and sell?
The BeeOmics Consortium sequenced the full genomes of over 1,400 colonies across Canada. Some colonies came from several different Canadian breeding programs which focused on bee health or on local adaptation. Other colonies came from well-known commercial breeders in Hawaii, New Zealand, and Chile.
This poster shows how different the genetics of different breeder stocks are, and also looks at how much genetic diversity there is within each stock. A companion talk by Rodney Richardson describes the impact of colony diversity on bee health and performance, while this poster describes specific genetic sources of that diversity. Our results will be of interest to beekeepers, queen breeders, and bee geneticists.

[P.02.37] Genetic diversity of *Apis dorsata* aggregation on Phung tree, Ficus albipila at Tanawsri mountain range, Thailand based on microsatellites

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The research aimed to study the genetic diversity of The giant honeybees (*A. dorsata*) from Tanawsri mountain range based on DNA microsatellites. The DNA samples of total 72 honeybee colonies from 9 aggregation trees were collected from Huai Sat Yai (Prachuap

Khiri Khan), Suan Phueng (Ratchaburi) and the Thai–Myanmar border along the Tanawsri mountain range. Five microsatellite loci (A14,A24,A88,A107 and B124) were used to study on population genetic of *A. dorsata* in Tanawsri mountain range. The results showed all study loci are polymorphic which had total 54 alleles. The highest number of alleles was 24 in A107 locus while, the lowest number of alleles was 3 in A24 locus. The genetic distance ranged 0.166 to 0.551. The neighbor joining tree revealed three group of the bees in which all of them are locality independent. This demonstrated that *A. dorsata* from the studied aggregation sites are panmictic population. Our results provide useful information for effective management and conservation of *A. dorsata* in Western Thailand, in particularly the aggregation population along of Tanowsir moutain range.

[P.02.38] Genetic diversity of Iranian honey bee (*Apis mellifera meda* Skorikow, 1829) populations via ISSR markers

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Identification and analysis of the genetic diversity of honey bee populations is one the most important goals in the honey bees breeding. For this purpose, a total of 88 young worker honey bees from Lorestan, Esfahan, Chaharmahal and Bakhtiari, Fars, Kerman and Sistan and Baluchestan provinces were collected and subjected to molecular analysis. In the present study, total DNA was extracted from the head and thorax sections of each honey bee worker, using salting out method with minor modifications. PCR amplification of genomic DNA was performed using 10 ISSR marker primers (including: A1, A2, A3, A4, A5, A6, A7, A8, A9 and A10). The PCR products were then subjected to 1.5% Agarose gel electrophoresis. The Shannon index and Nei gene diversity index were calculated using POPGENE V. 1.31, and the molecular analysis of variance (AMOVA) was estimated by GenAlex V. 6.3. Also, the polymorphism percentage, marker index (MI), Effective multiplex ratio (EMR), and Resolving Power (RP) were calculated. The highest and lowest Shannon index were observed in A1 and A9 primers, respectively. The average Shannon index ranged from 0.248 ± 0.288 to 0.356 ± 0.286. The highest and the lowest Nei gene diversity were observed in the A1 and A9 primer. The mean Nei gene diversity in the studied populations ranged from 0.166 ± 0.201 to 0.239 ± 0.214. The highest and lowest polymorphism percentage, marker index (MI), Effective multiplex ratio (EMR), and Resolving Power (RP) were observed in A1 and A9 primers, respectively. In this study, primer A1 had the highest polymorphism. So as the best primer for the study of genetic diversity honey bee populations is recommended. Moreover, according to molecular analysis of variance (AMOVA), intra-population and inter-population genetic diversity was estimated to be 35 % and 65 %, respectively. Creating queen rearing stations and implementation of interbreeding programs should be necessary because of considerable level of genetic diversity for given honey bee populations.

[P.02.39] Gene Expression Profiles in Honeybee Brains under the Food Search Box Protocol

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Honeybees are central place foragers. Part of what allows the bees to improve their foraging skills is their ability to learn where food patches are relative to the hive, avoid predation risk and navigate new locations. The molecular mechanisms underlying this learning ability have been investigated by using Proboscis Extension Reflex (PER) and free-flight learning protocols. Examples of genes which are up-regulated in response to visual and olfactory learning in honeybees include Early Growth Response protein 1 (EGR1), cAMP response element binding protein (CREB), dopamine receptor 1 (dop1) and dopamine receptor 2 (dop2). However, very little attention has been given to changes in gene expression during spatial learning, a key feature of navigation in honeybees. Here, we utilize a newly developed learning protocol called the Food Search Box (FBS) paradigm to study spatial learning in the honeybee. By conducting time-course experiments in the context of FBS, we are investigating how gene expression changes over time in response to spatial learning.

[P.02.40] The study of genetic diversity and phylogenetic relationships of Iranian honey bee (*Apis mellifera meda* Skorikow, 1829) populations via morphological characters

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Garmian University, Kalar, IRAQ

In the present study, in order to investigate genetic diversity and phylogenetic relationships among the Iranian subspecies honey bee populations morphological markers were used. Sampling was carried out during summer 2014 from 20 provinces of Iran (including West Azerbaijan, East Azerbaijan, Ardebil, Kurdistan, Zanjan, Gilan, Mazendaran, Golestan, North Khorasan, Razavi Khorasan, Hamedan, Kermanshah, Ilam, Tehran, Lorestan, Isfahan, Chaharmahal and Bakhtiari, Fars, Kerman and Sistan and Baluchestan provinces). Totally, 2250 young worker honey bees were investigated using 14 morphological characters (including: forewing length, forewing width, hind wing length, hind wing width, A4, D7, G18, proboscis length, cubital index, sternite index, scutellum color, third & fourth tergite length, hind leg length, and third tergite color). The results of variance analysis based on nested design and mean comparisons using LSD method showed that there were significant differences ($P<0.01$) between the provinces for all analysed morphological traits reflecting the existence of diversity between them. The results of the correlation coefficient analysis between morphological characters showed that significant correlation between most of the traits. Using Principal Component Analysis (PCA) 3 components were identified that explained more than

81.5% of the data variation. Cluster analysis using WARD method based on morphological characters classified samples in 2 groups. One of the groups involved the bees collected from North, Northwest and West parts of Iran, and the second group involved the bees which were collected from Eastern North, Central part and Southern regions of Iran. Also, a dendrogram based on WARD method divided 29 subspecies of the honey bee to 5 distinct clusters. The Iranian honey bee subspecies composed a shared clade with subspecies of Eastern Mediterranean, Near East and Eastern parts of the Middle East (O branches).

[P.02.41] Cuban honeybees: significant differentiation from European honeybees

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Historical antecedents of bee introductions as well as behavioral aspects of the bees have allowed to assume that Cuban bees are of European origin, and have remained on that way for approximately 60 years of isolation, even though the Latin America and the Caribbean region is practically Africanized. In this work, three bees were collected per apiary from 11 localities distributed throughout Cuba and their mitochondrial (by cytochrome b) and nuclear (STRs A8, A28, A43, A88 and A113) variation was characterized. Except for locus A28 ($p < 0.00$), none significantly deviates from the Hardy-Weinberg equilibrium, and as a whole showed the absence of population structure on the island (Fst = 0.059) and low consanguinity (Fis = -0.135). The Cuban population was compared with reference samples of European, Africanized and Brazilian bees and in all cases showed a moderate differentiation with respect to these populations (Fst paired of 0.156, 0.124 and 0.128 with Europe, Brazil and Africa, respectively). Although this differentiation, the population of Cuban bees shows a clear genetic affinity with the European population. Similarly, most of the samples have mitochondrial haplotypes of European origin (80%), while the rest corresponded to African haplotypes. In short, Cuban bees would have a European origin and would have differentiated in isolation, with low entry of bees of African lineages.

[P.02.42] BeeEducated: A Citizen Science approach to beekeeping. Helping new urban beekeepers off to a healthy scientific start!

T. Whittam
UTBA Urban Toronto Beekeepers Association, Toronto, CANADA

Citizen Science is defined as scientific research being performed by harnessing the power of ordinary people driven by curiosity. BeeEducated is a program of the UTBA (Urban Toronto Beekeepers association) to get new beekeepers off to a healthy start with their bees. Starting with the basics, BeeEducated covers everything a new beekeeper needs to know but with an emphasis on scientific published material. UTBA meets monthly and utilizes facebook and a webpage/blog to keep in touch with our 1400+ members. UTBA networks closely with the OBA (Ontario Beekeepers Association), OMAFRA (Ontario Ministry of Agriculture and Rural Affairs), OBA TTP Tech, transfer program at the University of Guelph. These organizations support our mission statement which is: Sustainable Beekeeping through Education and Mentorship in the greater Toronto region. Beekeepers are naturally curious. UTBA strives to base all our monthly education sessions on current accurate scientific fact rather than opinions. Sessions are interactive and encourage members to present their observations, measurements, statistics, trends, data and ask questions. Monthly meetings cover a wide range of topics: Bee biology, varroa, mite counts, treatments, equipment, pests, threats to pollinators, stings and allergic reactions, diseases and viruses, record keeping and hive inspections. Our apiary of the month session topic gives our members an opportunity to present their apiary setup and show us their data. The UTBA Facebook and webpage/blog allow us to reach out beyond the classroom to 1400 members and growing. BeeEducated is all about harnessing and molding citizen science beekeepers to observe, measure, analyze, report and share. The presentation if selected will elaborate on how BeeEducated is advancing, what we have learned and where we are going.

[P.02.43] Citizen Science with Open Data and Open Source Software: A Case Study

C. Poerschke
News Search Infrastructure, Bloomberg, London, UNITED KINGDOM

As a beekeeper and open source software (OSS) contributor, I was curious about the use of OSS in bee science and delighted to discover that a team of researchers at Freie Universität Berlin had published an open access sample dataset in 2018.
As described in their ''Tracking All Members of a Honey Bee Colony Over Their Lifetime Using Learned Models of Correspondence'' (https://doi.org/10.3389/frobt.2018.00035) open access paper, the team marked about 2,000 honey bees and continuously recorded them in a one-frame observation hive over 10 weeks with four cameras at a 3 frames per second sample rate. The recordings were subsequently analysed using computer vision and other algorithms to determine the bees' orientation and positions. The ''BeesBook Recording Season 2015 Sample'' (https://doi.org/10.7303/syn11737848.1) dataset is available online and covers 3 days extracted from about 3 million images.
Could I become a citizen scientist and do something interesting with this open dataset sample? As someone interested in both apiculture and computing, I dubbed my adventure ''Bee Informatics'' and followed where my curiosity and the data led. As and when time permits, I

share my investigations and code at https://github.com/cpoerschke/bee-informatics.
In this presentation, I'd like to:
(1) share highlights of my ongoing citizen science journey into bee data;
(2) summarise how collaborative distributed open source software development can work (based on my 5+ years of experience contributing to the Apache Lucene/Solr project); and
(3) sketch out, and perhaps inspire, ideas for citizen science hackathons or projects using open bee data.

[P.02.44] Ethics and beekeeping

F. Saucy
Société romande d'apiculture, Vuippens, SWITZERLAND

Ethics and beekeeping: The bee as a universal value for the 21st century.
As far back as we know, the honey bee is present in the collective imagination of human civilizations where it occupies a unique place. Considered as a deity in antiquity, the bee lost this special status with the advent of monotheistic religions. However, despite this relative decline, the bee has nevertheless continued to play an important role over the last two millennia as a source of sweet food and wax for lighting and religious rites. Until the Enlightenment, beekeeping, although widespread, remained relatively unproductive, more of a harvesting activity than an agricultural production activity. The situation changed drastically in the 19th century with the invention of the mobile-frame hive, which opened a real era of professional production and zootechnical practices typical of modern agriculture. The environmental crisis at the end of the 20th century, marked, among other things, by the decline of honey bees, wild bees, pollinators in general and losses in biodiversity, gives the bee a new and unique place in the community of threatened species, that of an umbrella species, an archetypal and sacrificial victim whose population status has become the emblem and flagship of ecological problems in the broad sense: protect the bee, is considered as a duty, as a means of survival of humanity. In this perspective, the bee has the role of universal value for the 21st century. From this observation, the ethical aspects of beekeeping will be critically examined, starting from considerations of philosophy and classical ethics, to go beyond anthropocentric views and lead to environmental ethics.

BEE BIOLOGY

10 SEPTEMBER 2019

POSTER SESSION 08 08:30-18:00
BEE BIOLOGY II POSTER AREA

[P.08.161] Toxin in nectar effect on bee olfactory learning in two cavity-nesting honey bees

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The thunder god vine, Tripterygium hypoglaucum, is a toxic nectar plant mainly range from southwest of China to northeast of China. A terpenoid, triptolide (TRP), in nectar could cause various behavioral damages on bees, including foraging responses, dance communication, and even olfactory learning. What will be the difference between the native species *Apis cerana* and the introduced species *A. mellifera* in react to triptolide nectar? We proposed two different hypotheses: firstly, *A. cerana* was sympatric with T. hypogiaucum, and after long coevolution it may have higher toxin tolerance than *A. mellifera*. Secondly, *A. mellifera* may have higher toxin tolerance than *A. cerana* because the larger size can withstand higher doses. Triptolide tolerance thresholds of these two bee species were determined with acute and chronic TRP exposure to them. When compared the acute effects of TRP on olfactory learning and memory between *A. cerana* and *A. mellifera*, the results showed that *A. mellifera* were more vulnerable to high concentration TRP sucrose syrup (5 and 10 µg TRP ml-1) than *A. cerana* did. The olfactory learning and memory of both *A. cerana* and *A. mellifera* were influenced, after one week pre-fed on TRP sucrose syrup. However, the concentrations of TRP contributed differently to affect these two bees species. The TRP tolerance thresholds of *A. cerana* almost 9 times higher than *A. mellifera*. Our results supported for which the native species *A. cerana* had higher toxin tolerance than the introduced species *A. mellifera*, after long coevolution. *A. cerana* may have evolved detoxification mechanisms that do not exist in allopatric bee species, *A. mellifera*. These findings have significant implications for understanding the coevolution of toxic nectar plants and pollinators and for how to the management of *A. mellifera*.

[P.08.162] Tracing the Fate of Pollen Substitutes in Honey Bee (*Apis mellifera L.*) Colonies

E. Noordyke, J. Ellis
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Commercial beekeepers need healthy, productive honey bee (*Apis mellifera L.*) colonies even when the landscape lacks adequate pollen forage to sustain the colonies. As a result, many commercial beekeepers spend significant money and labor on the use of pollen substitutes in their colonies. Despite this, there is little consensus in the literature about the benefits and drawbacks of protein supplementation on honey bee colony health. We aim to close part of this gap by investigating how honey bees distribute supplemental patties throughout their colonies. We traced the fate of three pollen substitute patties, commercially available in the U.S., throughout honey bee colonies by dyeing them with a non-toxic food coloring. Using spectrophotometry, we analyzed adult bee guts, larval guts, bee bread stores and colony debris (inside and outside of the hive) for presence of the dye. Our data show that a proportion of adult bees ingest the patty, as expected, though it is not clear if they utilize it or feed it to larvae/store it as bee bread. Understanding how honey bees utilize pollen substitutes will lead to better recommendations for beekeepers regarding the use of the supplements, especially in light of recent data that show mixed colony responses to these diets.

[P.08.163] Morphometry in oenocytes and trophocytes of the fat body in *Apis mellifera* larvae affected by "River Disease" in Uruguay

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The River disease is a clinical feature that affect honey bee colonies and is characterized by massive dead of larvae. The etiology of the colony intoxication is caused by the ingestion of secretions of Epormenis cestri dwelling located in trees Sebastiania schottiana near freshwater systems in Uruguay. The bee larvae fatty body tissue has oenocytes and trophocytes, considered the main metabolic cell of the bee larvae. There is no data referring the effects of River disease on the fat body cells, oenocytes and trophocytes, essentials in the larvae development. Therefore the objective of the present study was to determine whether toxic honeydews affects the morphology of this cells in larvae. Colonies were divided into a control group fed with common honey (n=5) and a group fed with honeydew from hives affected by River disease (n=5). Larvae were collected on day 5, fixed, and processed histologically for morphometry. The area, cellular diameter in oenocytes and trophocytes were measured. In larvae fed with toxic honeydews both cells, oenocytes and trophocytes showed loss of the typical spherical shape of their nuclei and cytoplasm, loss of heterochromatic nucleus, in trophocytes the presence of lipid vacuoles decreased. In contrast, the oenocytes of control larvae showed defined contours, acidophilus cytoplasm and euchromatic nuclei. Also the trophocytes of control larvae fed with honey had a rounded shape, lipid vacuoles and euchromatic nuclei. The oenocytes cellular area decreased in affected larvae (3274µm^2±147 versus 3933µm2±147; P=0.002). Comparable decrease were observed in oenocytes diameter (75µm±1.96 versus 86µm±1.96; P=0.0002). Likewise, trophocytes cellular area decrease in affected larvae (1348µm2±85 versus 2236 µm2±85; P<0.001). Consequently, trophocytes diameter decrease in affected larvae group (50µm±1.41 versus 65µm± 1.38; P<0.0001). In summary, the intake of toxic honeydews affected the morphology in cells oenocytes and trophocytes in bee larvae. These changes observed suggest that the River disease induce a deregulation of fatty body cells during bee larvae development.

[P.08.164] New tools to study nectar consumption in honey bees and Bumblebees: do bees follow economic laws of consumptions?

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We propose here a new patented device associated to new automatized laboratory protocols to measure the consumption of caged honey bees. Contrary to the standard protocol using free access to food, in our conditioning chambers, honey bees receive only a small amount of syrup (most often less than 1µl) when they visit the feeder. In other words, bees have to work a lot to get only small amounts of food.
Because we know exactly when bees eat, and what they get, with our protocols, it is easy to measure feeding patterns. Moreover, as we are able to control the access to the nectar resource, our tool is also perfect to study the determinants of food consumption. For economists, price is one of the most important variable that affect consumption. In that context, the economic demand law states that when price is increasing, consumption should decrease, and demand may be elastic, or inelastic.
During successive days, we increased the price of syrup and measured the daily food consumption of honeybees and *Bumblebee*s.
We discuss the results we gathered in the context of the demand law.
In conclusion, we will suggest some possible applications of such protocols and results to the study of pesticide effects.

[P.08.165] Effects of fenolic bioactive substances on reducing mortality of intoxicated bees (*Apis mellifera*)

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The study was focused on tests of biologically active substances (BAS) (mixture of fenols acids and flavonoids) on longevity of worker bees intoxicated by thiaclopride. Impact of BAS was tested in vitro under caged bees. Bees were split into four groups depending on the type of feeding. Each group contained 3 cages with 30 bees all at same ages. Negative control group (S) was feeded with sucrose solution (50% w/v). First experimental group (F) was feeded with sucrose solution (50% w/v) + mixture of fenols and flavonoids. Second experimental group (FT) was feeded with sucrose solution (50% w/v) + mixture of fenols and flavonoids + thiaclopride. Positive control group was feeded with sucrose solution (50% w/v) + thiaclopride (ST). Significantly higher mortality was found in group ST, whilst the lowest mortality was found in group S. The groups F a FT were very similar and statistically insignificant differencies between mortality. Decreasing of mortality in group FT in compare to the group T was probably caused by increased detoxification capacity. However, the increased mortality in group F in contrast to the group S indicates, that the use of BAS leads to increased metabolic load.

[P.08.166] Volatile compounds emitted by bee bread vary according to local climate and flora

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Honey bees preferentially consume pollen that has been stored in the comb and fermented; after being processed by the bees, it is called bee bread. The fermentation in bee bread, which reduces the pH, occurs with a progression of dominance of various species of microorganisms, especially acid-lactic bacteria. The profile of compounds found in each bee bread sample is a function of both the fermentation process and the plant species from which the pollen is collected. There is little information on the volatile components of bee bread; such knowledge would help explain why it is attractive to and preferentially consumed by the nurse bees. Identification of these compounds could aid in the development of protein diets that are readily consumed by bees. We collected bee bread from 46-48 hives located in each of two strongly contrasting environments in Brazil. These were Mossoro, Rio Grande do Norte state (5 degrees S latitude), with a hot semi-arid climate, and Porto Alegre, Rio Grande do Sul state (30 degrees S latitude), with a cool humid subtropical climate. The local flowering plant species are completely distinct in these two regions. The volatile compounds were analyzed with a combined gas chromatography–mass spectrometry apparatus. The bee bread samples from Mossoro produced 92 volatile compounds and those from Porto Alegre produced 60. In both locations, terpenes were the most abundant, comprising 29 terpenoids from Mossoro and 21 from Porto Alegre. Among these, 16 were exclusively found in the Mossoro samples and nine exclusively in the Porto Alegre bee bread samples. We conclude that though some volatile compounds are common to bee bread collected from colonies in distinct climate regions, others are exclusive to such regions. The differences are apparently due to variation in pollen sources, associated microorganisms and climate conditions.

[P.08.168] Effects of artificial diets on honey bee colony development during dearth period

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Honeybee Colony health is of great importance to the apiculture industry and to agriculture generally. The quality of diet influence colony health and strength, especially for colonies to survive harsh summer dearth periods in the region. Three protein rich pollen substitutes and supplements were formulated including: brewer's yeast, defatted soybean flour, parched black gram, skimmed powder milk, sugar cane molasses, date paste, sugar and honey. These formulations were feed to *Apis mellifera* colonies in the form of patties on top bars of the hive during summer dearth periods. Diet1 [brewer's yeast (35%) + defatted soybean flour (35%) + sugar (20%) + honey (10%)] was the most suitable with maximum 2041.2 cm2 sealed brood area, 5.5 covered frames. Honey produced was 92.36 % more than unfed colonies. Adult populations in colonies fed Diet1 didn't differ from those fed Diet2, and were significantly larger than colonies fed Diet3 and unfed ones. On the basis of preference of bee, it could be recommended to feed Diet1 or Diet2 to honeybee colonies during dearth period to improve the activities of honeybee colonies, for maintaining the strength and productivity.

[P.08.169] Consumption of protein patties and effect on spring population build-up

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Feeding protein supplements in spring is an integral component to colony build-up in preparation for honey production or pollination in Alberta, Canada. Beekeepers have access to many protein options, but little empirical research on their performance is available. In 2018-19, this project evaluated the consumption and impacts on colony build up of multiple protein patties containing pollen or pollen substitutes such as Feedbee, and Pollen-ate. In 2018, 120 single brood chambered colonies which had been overwintered indoors were fed one of four treatments: 1) Global 15% pollen patties, 2) Pollen-Ate patties, 3) Feedbee patties, or 4) trio patties (1/3 of each type of patty). Colonies consumed statistically more of the trio than the Global 15% pollen and the Pollen-Ate which were equivalent, and significantly less of the Feedbee patty than the other patties. Consumption rates of the three components of the trio patty were similar to the overall findings. No significant difference in colony build-up (cluster score or brood area) was observed between the treatments, but this may have been due to the late spring of 2018 when abundant natural pollen quickly became available. Results from a second trial conducted in spring 2019 will also be discussed.

[P.08.170] Evaluation of different pollen substitutes diets for Honey bee (*Apis mellifera L.*) during dearth period

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Honey bee (*Apis mellifera L.*) colonies face declines commonly attributed to pathogen, pesticide, parasite stress and nutritional deficiency. Commonly pollen substitutes diets were evaluated based on colony development and consumption. Here we examined three different proteinaceous artificial diets during dearth period by measuring area of sealed brood, pollen storage area, honey storage area and adult bee population. The highest area of sealed brood (157.6 cells/colony) as well as honey storage area was found in colonies fed with sucrose syrup with skimmed milk. The bees fed sucrose syrup with vitamin B-Complex syrup had their significantly higher bees pollution and honey storage area than control while sucrose syrup with soybean powder enhances sealed brood than control. Hence beekeepers are recommended to use rich proteinaceous diets during dearth period as a pollen substitute.

[P.08.171] Biological action of trace elements citrates on melliferous bees in different life periods

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The influence Co, Ni, Ag, Cu citrates obtained by the nanotechnology method on the mineral and lipid metabolism indices in the honey bees organism, their level in the products was investigated. Adding citrates of these mineral elements to the components of artificial feeding causes their corrective influence on the content of trace and macro elements, common lipids and their classes in the tissues of individual anatomical units and the entire organism of bees, as well as their products. The stimulating effect of the citrates of these elements is established on the intensity of the oviposition of the bee queens, which is more pronounced for Co and Ni in the spring period of living activity. The addition of Ag and Cu sugar and syrup citrates in doses of 0.2 and 0.5 mg resulted in a certain corrective action on the content of separate trace elements in the tissues of the honey bees of experimental groups and antagonistic effect on the level of such heavy metals as Pb and Cd. The content of Cu in the tissue homogenate of the bees' organism of the experimental group decreased by 2.4%, the level of Zn - by 18.8%, compared with the control against the background of growth of Cr and Co concentrations, respectively, by 26.7% and 53% ($p < 0,05$) .

Comprehensive feeding of honey bees with citrates of trace elements in different dilutions with sugar syrup caused corrective effect on mineral metabolism with an increase ($p < 0,05$-$0,001$) of Co, Zn, Cu and Ni content in the tissue homogenates of the entire organism.

The conducted investigations showed that both separate and joint addition of trace elements citrates to honey bees in the spring period led to evenly directed changes in the content of separate mineral elements in their tissues. These interactions between mineral elements must be taken into account in the schemes of mineral feeding of bees in spring. Activating the physiological capabilities of honey bees by feeding organic salts of trace elements is a safe alternative that can be used to improve the viability and productivity of bee families in certain seasonal periods.

[P.08.172] Effect of protein supplementation on honey bee drone rearing, morphology and semen quality

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The objective of this study was to analyze the effect of feeding honey bee colonies a commercial protein supplement on drone rearing and morphology, as well as on the quality of their semen. Four colonies were randomly assigned to the supplement and sugar syrup treatment and fed bi-weekly for five weeks. The control group was fed only sugar syrup. The number of drones reared was determined and when they reached sexual maturity, samples of them were collected and the quality of their semen assessed. Significantly less drones were reared in the colonies fed with the protein supplement than in the control colonies ($p < 0.05$). However, the drones reared in the protein-supplemented colonies weighed significantly more (10.5%) and had a higher thoracic index than drones reared in the control colonies. Additionally, the supplement increased most of the variables used to assess semen quality, including sperm concentration ($p < 0.001$) and viability ($p < 0.05$). Abnormalities in sperm morphology included spermatozoa with large heads, rolled tails and fragmented tails. A lower percentage of anomalies were found in the semen from drones reared in the protein-supplemented colony. The minerals found at higher amounts in drone semen and seminal vesicles were selenium, magnesium, copper, zinc, calcium and phosphorus. It is concluded that feeding a commercial protein supplement to drone-rearing colonies improved drone morphology and semen quality, but not drone production.

[P.08.173] Development of melezitose in honeydew and its impact on honey bee health

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Cement honey is a major issue for beekeepers, representing a special kind of honeydew honey with high levels of the trisaccharide melezitose. Honeydew honey is the result of insects (Hemiptera) sucking phloem sap of trees and excreting the sugars in honeydew droplets. Subsequently honey bees take up the honeydew and process it into honeydew honey. Honey with high amount of melezitose crystallizes and obstructs the combs, leading to an economical loss. Nevertheless, precise analyses of the conditions under which melezitose occurs have not been realized. Furthermore, it is not known which impacts the trisaccharide has on honey bee health.
In order to determine influence factors for the emergence of melezitose, more than 600 honeydew droplets from defined species were collected under different environmental conditions and the sugar spectra were analyzed by high performance anion exchange chromatography with pulsed amperometric detection. To obtain the impact of melezitose on honey bee health, additional feeding experiments with daily evaluation of food uptake and mortality have been realized. Additionally, comprehensive 16S rRNA Illumina sequencing of the gut microbial community has been performed.
We found remarkable differences in the amount of melezitose between honeydew collected from different honeydew producer species and according to different environmental conditions. Bees fed with melezitose showed increased food uptake and higher energy consumption than control groups. Furthermore, melezitose feeding caused disease symptoms such as swollen abdomen, abdomen tapping and difficulties with movements and a significantly higher mortality than in control groups. Gut microbiota analyses indicated a shift of the lactid acid bacteria in melezitose fed bees.
Our study provides important information, under which conditions melezitose will be produced and therefore how the occurrence of cement honey can be avoided. Additionally, feeding experiments indicate the high effort that is required for the degradation process of the large-molecule melezitose. This effort might lead to a higher intake of food and a higher susceptibility to intestinal diseases. Finally, we present evidence that the gut microbiota is significantly involved in the digestion of melezitose.

[P.08.174] Supplementation containing probiotic and prebiotic increases expression of Africanized *Apis mellifera* major royal jelly Protein-3 mRNA

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Royal jelly is a bee product widely studied as to its composition in physiologically active substances, allowing the differentiation of a young

larva, from a fertilized egg, into a queen and / or initially nourishing a worker's larvae. Royal Jelly has protein in approximately 50% of its dry mass. Among the proteins we highlight Major Royal Jelly Proteins (MRJPs), especially MRJP3. Our research group (GPBEE) has been developing selection of queens producing royal jelly that have already known and selected Mrjp3 genotypes. In this study Africanized *Apis mellifera* workers were evaluated for expression of Mrjp3 messenger RNA after supplementation with prebiotic and/or probiotic containing diets. The experiment was started at the Experimental Farm Iguatemi-FEI, Universidade Estadual de Maringá (UEM), Maringá, Paraná, Brazil. Newly emerged bees were kept in experimental cages for a period of 11 days. The treatments were: base protein supplement (RPB); prebiotic and probiotic protein supplementation (RPP); negative control containing pollen, sugar syrup and water (-) and positive control containing both types of supplement (+). During the experiment three collections were carried out. The laboratory analyzes were carried out in the Department of Biotechnology, Genetics and Cell Biology - UEM. Using the qRT-PCR technique it was possible to analyze the mean values of $2^{-\delta CT}$ of the mRNA transcripts of the Mrjp3 (Mrjp3 mRNA) gene according to the time of treatment and the different types of supplementation. In the collections of workers with four and 11 days, the lowest expression values of the Mrjp3 mRNA were observed. The highest expression values of Mrjp3 mRNA were detected in workers with seven days. At this age we observed that there was a 6.8-fold increase in the expression of Mrjp3 mRNA when they were treated with (+), ie the two types of supplement; the RPP treatment promoted a 5.1-fold increase in Mrjp3 mRNA and RPB increase of 1.8-fold. The use of protein supplementation containing prebiotic and probiotic possibly promote an increase in the expression of the Mrjp3 gene, contributing to the production of royal jelly. Therefore, commercial use of this supplement may be promising for beekeepers feeding royal jelly bees producer.

[P.08.175] Honey Bee Nutrition - Early detection of malnutrition and colony collapse

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The nutritional status of managed honey bees depends on the attention that beekeepers provide them and their hive management skills. One major issue beekeepers have to deal with, is the early detection of what is termed the skinny bee syndrome, where bees suffer nutritionally and become so unwell that they cannot recover easily. At this stage, the colony is unable to continue foraging and support their brood, commonly by the time this is detected it is too late for beekeeper intervention. The provision of pollination services and more over the production of mono-floral honey are valuable incomes for the Australian honey bee industry. Therefore, it is essential for beekeepers to maintain their bee colonies in a healthy nutritional condition for optimal productivity. Here we identify the time point when honey bees are on a nutritional decline and determine the best nutritional management strategies to minimize the risk of productivity loss. Using Liquid Chromatography-Mass Spectrometry (LC-MS) metabolomic profiles of the bees at different time points of nutritional starvation and supplemental feeding were compared. We identified differences in amino acids, lipids, carbohydrates and proteins expression that reflect changes in the bee's physiology. Finally, the molecular profiles could be correlated with hive condition indicators; such as brood and honey quantity, hive weight, temperature and humidity.

[P.08.176] Comparison between tradition and industrial production of Caucasian honey bee queen

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The effect of inclusion of olive oil and flaxseed oil in the pollen substitute on egg laying rate, population, body weight and body fat content of honey bee.
The present study was carried out to evaluate the effect of olive and linseed oils in pollen substitutes on queen's laying, colony population and honeybee body weight. 24 hives were tested in this study in a completely randomized design with 3 treatments (8 hives per treatment). Treatments including control ration (including honey, sugar and pollen), ration 2 (including pollen substitute cake made from honey, sugar, olive oil) and ration 3 (including pollen substitute cake prepared with honey, sugar, linseed oil). The duration of the experimental period was 60 days, which consisted of 14 days of adaptation and 46 days of the main test. During the experiment, bee colonies in experimental hives were fed with pollen and pollen substitutes in different experimental treatments. A plastic rectangular paper with 1 × 1 inch standard square covering all of their parts was used to measure the queen's laying every 7 days. The protein content of the honey bee body and their pupae was measured by the Kjeldahl and fat content measured by Soksele method. The results of this study showed that the use of pollen substitutes increased the feed intake and the body weight of the tested honeybees significantly ($P < 0.05$). Pupae and honeybee protein content were not affected significantly ($P > 0.05$). The pupae fat content of olive oil group (2.90%) and linseed group (1.96%) was higher than control (1.63%) group ($P < 0.05$). The bees fat content were not affected significantly ($P > 0.05$). The bee population of olive oil group was higher than other groups ($P < 0.05$). Also, the queen laying were not affected significantly ($P > 0.05$). To sum, it can be declared that olive and linseed oils can be used as the main ingredients of pollen substitutes.

[P.08.177] The pollen of pyrostegia venusta (bignoniaceae) non-influence in the longevity of honeybees in controlled conditions

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Pyrostegia venusta, a plant of the family Bignoniaceae, is widely distributed in Brazil. Studies have revealed that this plant is toxic for some animals, such as cattle and Artemia salina, for instance. For this reason, the possible effect of Pyrostegia venusta pollen toxicity on the longevity of the Africanized bees, *Apis mellifera*, was investigated. Combs of three distinct colonies containing broods close to emerge were collected, and taken to an incubator with controlled temperature at 35°C until the emergence of bees. A total of 50 newly emerged bees were then transfer to wooden cages measuring 8 x 11 x 13 cm and kept in a BOD incubator with controlled temperature at 35°C. Group 1 (control) was supplied with basal diet food, a commercial suplement known as MegaBee®, considered a nutritional supplement containing minerals, lipids, vitamins and carbohydrates; group 2 (treatment), had pollen of P. venusta added to the basal diet (MegaBee®) at concentrations 0.5%, 0.75%, 1.0% and 1.25%, and offered to the bees. Bee mortality was daily measured to evaluate the effect of a possible toxicity of pollen provided as diet. Results showed that the bees from control group presented a higher survival rate, 19 days, while bees from the treated groups (0.5%; 0.75%; 1.0% and 1.25%), survived 13, 15, 16 and 15 days respectively. The Kaplan-Meier Survival Analysis: Log-Rank, showed no statistical difference ($P = 0.214$), revealing that the longevity of honeybees were not influenced by a possible toxicity from the pollen of P. venusta. Thus, it is possible that the toxicity of P. venusta for other animals, is associated to pyrostegine, a glycoside present in the plant leaves.

[P.08.178] Effects of Different Nutrition Levels of Chlorella Alcohol on Laying the Queen Bee Colonies of *Apis mellifera*

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In order to evaluate the nutritional effects of various levels of chlorella algae as a protein supplement on laying queen bee rate, larvae and pupae, This experiment were conducted based on Completely Randomized Block Design (CRBD) with 4 treatments and 10 replications in wild land in Alborz province on (*Apis mellifera*). First, the colonies were identical in terms of honey, population, and queen sorority of the same age. Treatments: 1. Control diet was based on soybean, corn gluten, honey, pollen and sugar. 2. Raw diet is containing 5% chlorella. 3. Raw diet is containing 10% chlorella. 4. Raw diet is containing 20% chlorella. The Comparison between experimental treatments showed that diets had no significant effect on the amount of oviposition and larvae. However, the difference was significant for pups ($p < 0.05$), the highest percentage of pupae belonged to treatment 3 (2644.33 centimeters) and the least belonged to treatment 2 (2070.58 centimeters). There was a significant difference between experimental diets for breeding (total eggs, larvae and pupae) ($p < 0.05$). The highest rate of larvae was for treatment 3 and the lowest was for treatment 2. It seems that the nutritional effects of chlorella algae on infant fertilization are positive and can increase egg, larvae and pupae in comparison with the control group.

[P.08.179] First European study of honey production in beehives under an annual Vita Feed nutritional protocol

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In recent years, climate change, crop spraying and monoculture are some of the factors that have led to a decline in honey production worldwide. Variations in the content of available proteins exerts a serious effect on the needs of bees.

Five years of making records and evaluations in Argentina showed that supplementation with VitaFeed Nutri by sprinkling during the honey flow generated an average increase of 2.5 kg of honey per beehive. This led to the creation of an annual nutritional supplementation protocol with the objective to observe the impact on production of those beehives.

The 2014/15/16 seasons showed an average increase of 20% of honey for the colonies treated under the protocol, compared with untreated control colonies, so it was decided to repeat the test in order to confirm if we can achieve the same excellent results in Europe.

The tests were conducted in the 2017 season, at one conventional apiary located in Neo Klima town, in the island of Skopelos, Greece.

The apiary was divided into two groups, one group subject to the supplementation protocol and the second comprising control (only sugar syrup).

The nutrition plan was administered to the experimental colonies, consisting of: 300g of 'Vita BeeFood', 25 ml of 'VitaFeed Power' in sugar syrup and 50g of 'VitaFeed Nutri', the latter sprinkled on top of the brood frames.

The measurements were performed in the extraction room. From the field, honey supers were marked to extract honey were carried separately.

In this test in Europe, the honey production obtained was an average of 10.3 kg for the treated colonies and 7.71 for the control hives. This increase of 34% per hive not only shows the necessity of good quality of nutritional products but also show that a good investment has quick positive results. The use of products designed exclusively for bees shows a positive effect, which correlates with the last 5 years of trials carried out in Argentina.

[P.08.180] Regulation of the intake of dietary protein and fat by adult worker honeybees

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Honeybees collect and eat the floral pollen of many plant species to obtain protein and fat. Pollen of many plant species is mixed together to form 'bee bread' within a colony, which is then consumed by adult nurse bees who feed the larvae and the queen with glandular secretions. How nurse bees regulate their intake of protein and fat simultaneously during feeding has been seldom studied. Here we use the principles of the Geometric Framework for Nutrition to study the simultaneous regulation of protein and fat in diet by newly-emerged, adult worker honeybees. We used diets composed of essential amino acids (EAA), lecithin (F), and sucrose. We measured the food consumption and survival of bees that had either been confined to eat a specific ratio of EAA-to-fat (EAA:F) or gave bees the choice of diets with different ratios of EAA:F. In both assays, we found that adult workers regulated their intake around a value of 1:2. Bees survived well on all diets, even diets containing as much as 1:12 EAA:F. Bees fed with diets high in fat were more likely to have larger hypopharyngeal glands and to gain body fat. These data indicate that young adult worker honeybees require diets relatively high in fat compared to other bee species and other insects.

[P.08.181] Health of *Apis mellifera* bees in hot climate regions

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The objective of this research was to evaluate colonies of *Apis mellifera* bees for resistance to Nosema and Varroa destructor in the Agreste and Zona da Mata regions in Pernambuco, Brazil. The average annual temperature in Agreste and Zona da Mata is 23.8ºC and 25ºC, with annual rainfall of 640mm and 2,000mm respectively. Samples from 63 colonies were harvested. 20 colonies from the Agreste region and 43 colonies from the Zona da Mata region. The harvested bees were stored in a plastic container containing 70% alcohol. The tests were carried out at the Laboratory of Genetics and Breeding of Bees in the Department of Animal Science at the Federal Rural University of Pernambuco (DZ/UFRPE). For the Nosema analysis, the abdomens were removed from 25 adult bees, macerated with 25mL of distilled water and then filtered. The spore count was done in 5 quadrants and the estimated degree of infection was based on the Cornejo and Rossi (1975) scale for millions of spores per mm3. To determine the rate of infestation of Varroa per colony, approximately 200 adult bees were transferred to a beaker containing 200 ml of alcohol and water solution (1:1 ratio). The beaker with the bees was agitated and the contents transferred to a white container for the removal and counting of bees and mites. The methodology of De Jong and Mantilla (1986) was used to determine the infestation rate per colony. Data were analyzed using the chi-square distribution ($P<0.05$). From the results, it could be observed that 54% of the colonies did not present Nosema spores, 35% presented them in very light scale and 11% in light. From the Varroa destructor analysis a mean of 3.44 ± 4.69 was observed. The means of the infection rate with Nosema and infestation with Varroa in the two regions did not differ statistically. The colonies of *Apis mellifera* from the Zona da Mata and Agreste regions of Pernambuco present low rates of infection and infestation and are resistant to Nosema and Varroa destructor.

[P.08.182] Possible existence of a Varroa resistant honey bee population in Wales, UK

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The ectoparasitic Varroa mite (Varroa destructor) functions as a vector for the spread of bee viruses within and between colonies of the Western honey bee (*Apis mellifera* sp.), making it a driving factor behind honey bee colony losses. However, it is known there are populations of Western honey bees which developed traits to cope with the challenges presented by Varroa after being left unmanaged for this parasitic mite. During the last two decades, several beekeepers in North Wales – specifically in the Gwynedd county, including the LLyn Peninsula – no longer treat their honey bee colonies against Varroa. Interestingly, the local beekeeping community also does a lot of effort to prevent imports of honey bee colonies from outside the region which has allowed natural selection to occur for a more or less closed population. We monitored and compared colonies from both the treatment-free and a treated population in North Wales to locate traits contributing to resistance.

BEE BIOLOGY

11 SEPTEMBER 2019

POSTER SESSION 14 08:30-18:00

BEE BIOLOGY III POSTER AREA

[P.14.300] Identifying honey bee bio-markers in microbiome and proteome using machine learning

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Gut Microbiome and host health are often found to be associated; and recent works suggests analogous behaviour in honey bees. 16S Intestinal gut microbiome and host proteomics from 212 honey bee colonies across Canada were analyzed. Full microbiome and proteome data were processed through a machine learning analysis algorithm in order to narrow down the 5420 proteomic and microbiome features to a much smaller set of biomarkers with predictive strength. The resulting model was capable of predicting 10 different phenotypic and pathogen load variables (namely: Location of origin, hygienic behaviour, honey production, weight loss/gain, defensive behaviour, *Lotmaria passim* loads, nosema loads, varroa loads, bqcv loads, sbv loads) with an accuracy of over 90%, while narrowing down the dataset to using only 60 features from the initial 5420. This analysis demonstrates the possibility of using machine learning in combination with multiple dense data sources in order to discriminate key biomarkers to identify various host health conditions.

[P.14.301] Effects of Probiotic and Oxalic Acid on Honey Bee Microbiota and Innate Immunity

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University of British Columbia - Department of Biochemistry and Molecular Biology, Vancouver, CANADA

The honey bee gut microbiota contributes significantly to the health of its host and plays a vital role in stimulating the production of pathogen-fighting antimicrobial peptides. As we discover more about the importance of gut bacteria for innate immune function, interest in probiotics is growing and several products are appearing on the commercial market. But few studies have confirmed that candidate probiotics actually achieve the advertised health outcomes by changing the composition of the gut microbial community. Here, we examined changes in the composition of the microbiota after treatment with 1) a commercial probiotic, SuperDFM and 2) oxalic acid, which is an unrelated but common in-hive treatment for Varroa destructor. Effects on the immune system could be a result of changes in the microbial community composition or a direct result of the treatments themselves. Caged honey bees were treated with probiotic or oxalic acid before whole guts were subjected to 16S rRNA sequencing to determine the composition of the bacterial gut community. Then, antimicrobial peptide levels were determined by label-free, LC-MSMS-based quantitative proteomics. We will discuss the microbiome sequencing results and relate this to quantities of antimicrobial peptides as markers of innate immune system suppression or stimulation.

[P.14.302] Magnetic sensing and genes screening associated with light and magnetic field by RNA-seq in *Apis mellifera*

W. Yan, F.-Y. Zhao, X.-B. Wu

Honeybee Research Institute, Jiangxi Agricultural University, Nanchang, CHINA

The selection rate of forager bees for the presence or absence of a magnetic field, 2 concentrations of sucrose solution, and positions was analyzed by magnetic field sensing device. The RNA-seq technique was used to sequence the forager bees in both light and magnetic stimulation treatment (T1), only magnetic stimulation treatment (T2), and the non-light and magnetic stimulation treatment (CK). The KEGG analysis was carried out after searching the *Apis mellifera* 4.5 database and screening differentially expressed genes. Gene HACD,Hsp70,Hsp90,CRYAB,CPEB,GST,Bip,GRP94,PDE and GlyA were selected randomly for verification using qRT-PCR technology. The magnetic sense results showed that forager bees have direct preference in the target with the magnetic ($P<0.05$) and with 50% sucrose during the previous discriminative training ($P<0.05$). There is no preference in position ($P>0.05$). Results suggested that 1250 genes were obtained from T1,T2 and CK transcriptomes. KEGG assignments were used to classify the functions of the 1250 genes, and which could be categorized into 5 catrgories, Genetic Information Processing (26.01%), Environmental Information Processing (8.48%), Organismal Systems (3.08%), Metabolism (56.65%), Cellular Processes (5.78%). Combining with KEGG, 9 genes screened were associated with light and magnetic field. The results of qRT-PCR showed that HACD, Hsp90, CRYAB, Bip, GRP94, PDE and GlyA genes was consistent with the sequencing results of transcriptome. Forager bees of *Apis mellifera* can sense the magnetic field. The transcriptome results also provide a basis for further investigation of genes related to light and magnetic induction.

[P.14.303] Understanding consumer beehaviour: Using Phantom alternatives to nudge bee decision making

C. Forster, T. Latty, R. Gloag, D. Hochuli
The University of Sydney, Sydney, AUSTRALIA

Experiments on humans suggest that desirable but unavailable items (such as 'sold-out' options) can change, or even reverse, a consumer's preferences for other items. Desirable but unavailable items are known as 'phantom alternatives' and can be used to 'nudge' consumers into buying target products. Like a human consumer, bees foraging for flowers are faced with an overwhelming number of choices. Here we investigate the effect of phantom alternatives on the choice behaviour of bees. Bees were presented with either a binary choice set containing two equally preferred feeder types or a trinary choice set containing the target, the competitor and a desirable – but unavailable- phantom alternative. Just like human consumers lured in by items later revealed to be sold-out, bees altered their preferences when an attractive, but ultimately unavailable, item was added to their choice set. Our results show that the phantom alternative effect is not exclusive to humans, and we suggest possibilities for using the decoy effect to 'nudge' bee preferences in agricultural systems.

[P.14.304] Development of mass multiplication technique for stingless bee, *Tetragonula iridipennis* queens

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An attempt was made to raise Stingless bee, *Tetragonula iridipennis* queens in invitro condition at college of Forestry Ponnampet, Karnataka, India. To provide optimum conditions at the incubator, the temperature and relative humidity of the natural colony was measured by taking the observation for a period of two months. The mean temperature and relative humidity of the natural colony was 28.06±0.34 0C, 78.40±2.22 per cent respectively. The mean quantity of larval food in the worker cell was 2.8±0.15 µl. Double the quantity of this (5.6 µl) was provided to the brood invitro. The egg and larval stages were placed in effendorf tubes with larval food and temperature, relative humidity was maintained in the incubator similar to that of natural colonies except for first three days where in 100 per cent humidity was maintained. Out of the 48 eggs put into in vitro condition 11 developed to the larval stage, but then onwards failed to reach pupal stage. Out of the 48 larva reared in vitro, 16 completed the larval stage and 8 emerged as adults. The experiment showed the possibility of rearing T. iridipenis queen, a first attempt in this species of bees.

[P.14.305] Honey production and storage pots for the stingless bees *Tetragona clavipes* (Fabricius) and *Scaptotrigona bipunctata* (Lepeletier) in State of Paraná, Brazil (Hymenoptera, Apidae, Meliponini)

J.E. Melo Nascimento [1], F. Freitas de Oliveira [2], A.J. Souza Pacheco Filho [3], L.G. Sousa Perugini [1], W. Guadalin de Oliveira [1], G.H. Simões Pereira [1], V. Arnaut de Toledo [1]
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These bees are important as an alternative source of income for beekeepers who marketing their products and for pollination. This research was carried out to evaluate the colonies productivity of *Tetragona clavipes* (Fabricius, 1804) and *Scaptotrigona bipunctata* (Lepeletier, 1836), grown in the southern region of Brazil due their high productive potential and resistance to periods of food shortage. For this, we evaluated the honey productivity in five colonies of both species, using 10 pots as reference, based on the following parameters: height, diameter, volume and thickness of the honey storage pots. For the statistical analysis, the Mann-Whitney test (U) was performed due the non-normality of the data. T. clavipes presented significantly higher median values of height, diameter, thickness, volume and productivity per colony than S. bipunctata (height: T. clavipes - 50.79 mm, S. bipunctata - 25.50 mm, $U = 147$, $p < 0.001$; diameter: T. clavipes - 25.32 mm, S. bipunctata - 15.48 mm, $U = 248.5$, $p < 0.001$; thickness: T. clavipes - 0.5400 mm, S. bipunctata - 0.4650 mm, $U = 856.5$, $p < 0.001$; volume: T. clavipes – 15.0 mL, S. bipunctata – 3.0 mL, $U = 161.5$, $p < 0.001$; productivity per colony: T. clavipes – 900 mL, S. bipunctata – 330 mL, $U = 540$, $p < 0.001$).Although both species belong to the same tribe, both having quite populous and defensivenests, probably, the differences in the structureof honey pots (length, diameter, volume and thickness) interfered directly in the storage capacity of the colony and, therefore, in the productivity. Thus, if beekeepers from State of Paraná want to opt for a more productive species, although both species are excellent honey producers, T. clavipes presents a higher productive capacity, reaching a little more than 2.7 times the productive capacity of S. bipunctata. However, other parameters must still be evaluated as taste, texture and acceptance of the honey, to guide the decision of the beekeepers on which of the two species must be more relevantas a generation of income to the producing families.

[P.14.306] Trophic resources collected by stingless bees in the Brazilian Amazon

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In the Amazon region stingless bees represent a diverse group of social bees and are very important as pollinators. Understanding about the plants used as trophic or nesting resources by these bees may contribute to conservation of the tropical forests. Studies carried out in different localities of this region show the diversity of plants that are visited by stingless bees (Meliponini). A total of 47 pollen types

were identified in 22 botanical families in honey samples collected directly from the honey storage pots from colonies of Meliponini in the Nova América community of the Sateré Mawé tribe, located on the Maruá river in the municipality of Maués, Amazonas. The most common bees in this study were of the Melipona seminigra species with 31 pollen types collected, followed by Melipona sp. with 27 pollen types, *Scaptotrigona* sp. with 25 pollen types, and Melipona dubia with 19 pollen types. The most important families, were: Anacardiaceae, Arecaceae, Burseraceae, Chrysobalanaceae, Dichapetalaceae, Dilleniaceae, Euphorbiaceae, Fabaceae, Lecythidaceae, Melastomataceae, Myrtaceae, Rhamnaceae and Salicaceae. Analysis of pollen collected by Melipona (Michmelia) seminigra merrillae and Melipona (Melikerria) interrupta which are the two main species used in the beekeeping industry within the Central Amazon region, show a predominance of pollen from Fabaceae, Melastomataceae, Myrtaceae, and Anacardiaceae in the bee diet from várzea (floodplains).
Studies on plants collected by stingless bees will contribute to improve beekeeping in the Amazon allowing an economically important complementary activity in this region.

[P.14.307] Climatic and latitudinal effects on beekeeping, meliponiculture and bee colony losses in Latin America

F. Requier [1], K. Antúnez [2], L.A. Garibaldi [1], C.L. Morales [3], J.M. Rosso Londoño [4], A. Giacobino [5], M. Porrini [6], R.A. Velarde [7], M. Basualdo [8], Colony Losses Group [9]

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Large-scale monitoring programs applied to bees have allowed researchers to pinpoint the effects of climate change on the current patterns of decline in wild and managed bee populations in the United States and across Europe. Consequently, detailed knowledge on the bee decline patterns was restricted to these latitudes and to the specific climatic and environmental contexts of the Northern hemisphere. We performed the first large-scale, volunteer-based survey, to monitor honey bee and stingless bee (Tribu Meliponini) colony losses across Latin America. We aimed to describe the geographic patterns of bee mortality and explore the climatic, health and management drivers underlying that spatial heterogeneity. More than a thousand producers (893 beekeepers and 115 meliponicultors) participated of the survey during 2016-2017. The spatial scale of the responses included the complete latitudinal gradient of Latin America, i.e. from Mexico to Argentina, encompassing a broad range of climates and altitudes. While meliponiculture activities (the keeping of colonies of stingless bee species) were biogeographically restricted to sub-tropical and tropical regions with high rainfall and high temperature, beekeeping (the management of Western honey bee *Apis mellifera* colonies) was widely distributed over the climatic range. Interestingly, honey bee and stingless bee colony mortalities were not homogeneously distributed across the region. Here, we will discuss our results in relation to various potential drivers of colony loss that could explain the patterns we uncovered. We will specifically test the respective effect of environmental variables (climate, altitude, landscape), operation size, disease incidence (two well-established drivers of bee colony mortality in the Northern Hemisphere), and their interactions. We will be interested in exploring how the already identified drivers of colony loss (e.g. operation size, disease incidence) are representative in this region or either new drivers emerge due to unique characteristics of the data context (e.g. climate, altitude, landscape). These results will help our understanding of the processes involved in bee colony mortality, as well as provide tools for risk assessment for apiculture and meliponiculture.

[P.14.308] Healthy Bees™ - A Unique Solution To Providing Balanced Nutrition For A Sustainable Global Beekeeping Practice

F. Del Vecchio, L. Rosen, J. Turpin, A. Festuccia
Healthy Bees, Llc, Bay Harbor Islands, USA

Beekeeping practices and agricultural systems have experienced great change in the last several decades. This, along with the many stressors affecting managed colonies make feeding bees quality, balanced nutrition of paramount importance. Balanced nutrition not only allows for proper and efficient physiological development but can also increase immune function and help honey bees better deal with the pests and pesticides they may encounter.
In this talk, we will compare and analyze the use of our Healthy Bees™ supplement to existing supplements in terms of micro- and macro- nutrient quality, as well as how it affects the health of the hive. We will also show international and independent studies on how the balanced amounts of protein, amino-acids, minerals, carbohydrates, fats, vitamins, and especially antioxidants contained in our Healthy Bees™ supplement helps bees deal with oxidative damage caused by pesticide exposure, all of which suggests that Healthy Bees™ increases a bee's ability to detoxify toxins as well as fight infections. It has been proven that our product contains not only an average of 30 times more antioxidant power than other products for bees on the market, but tests and research have also shown that Healthy Bees™ performed stronger than vitamins B6, B12 and beta-carotene combined.

USDA studies showed, for the first time ever, that a nutritional system (Healthy Bees™) could impair the amount of ROS formed by herbicides, demonstrating that Healthy Bees™ can lower the detrimental effects of pesticides as well as boost the immune system of honey bees. This is a huge development in improving honey bee health in the field. Under Healthy Bees™ balanced nutrition, bees expressed fewer of the many genes responsible for the detoxification system, a strong indication that Healthy Bees™ was helping to detoxify the bees' systems.
We will discuss how soy- and corn-based supplements may be harming our hives, both in terms of nutrition and in terms of the agricultural system it supports. The full set of independent tests will provide evidence of why Healthy Bees™ will be the future of nutrition for our bees while supporting a new, sustainable global beekeeping practice.

[P.14.309] The Effect Of Carbohydrate Sources: Sucrose, Invert Sugar And Components Of Manuka Honey On The Composition Of Bacteria In The Digestive Tract Of Adult Honey Bees

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Bacteria within the digestive tract of adult honey bees are likely to play a key role in the digestion of sugar-rich foods. However, the influence of diet on honey bee gut bacteria is not well understood. During periods of low floral abundance, beekeepers often supplement the natural sources of carbohydrate food that honey bees collect, such as nectar, with various forms of sugars such as sucrose and inverted sugar (a mixture of the monosaccharides glucose and fructose). The effect of these sugar supplements on the abundance of gut bacteria in bees from a single colony were compared with the effect of two natural dietary treatments: manuka honey, a monofloral honey with known antibacterial properties, and the diet that bees consume in a hive. Sucrose solutions containing synthesised compounds associated with the antibacterial properties of manuka honey were included as an additional treatment. 16S ribosomal RNA (rRNA)-based sequencing showed that dietary regimes containing manuka honey, sucrose and invert sugar did not alter the abundance of core bacteria after 6 days. However, sucrose-rich diets increased the numbers of less abundant bacterial taxa, which significantly altered the bacterial composition. Acetogenic bacteria from the Acetobacteraceae and Rhizobiaceae families increased three-fold when bees were fed sucrose. These results provide evidence that sucrose fuels the proliferation of specific low abundance primary sucrose feeders, which metabolise sugars into monosaccharides, and then to acetate.

BEE HEALTH

09 SEPTEMBER 2019

POSTER SESSION 03 08:30-18:00
BEE HEALTH I POSTER AREA

[P.03.45] The impacts of developmental multi-pesticide pollen and wax exposure on queen honey bee (*Apis mellifera*) health, mating, and colony development

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Honey bee (*Apis mellifera*) queens are challenged by various developmental exposures which have the potential to not only influence individual queen survival but also downstream colony health. In this study, we reared queens in exposure environments containing realistic pesticide residues and examined the impacts of developmental exposure on queens and their established colonies. We used pesticide treatments consisting of a mixture of insecticides, fungicides, and herbicides that have been frequently detected within commercial colonies, with the toxicity of each mixture based on field relevant Hazard Quotients. We raised queens in simulated exposure environments by grafting worker larvae into queen cells made of treated or untreated beeswax and then rearing queens in cell builder colonies fed a pollen supplement with or without an added pesticide treatment. We later established queens into full size colonies and recorded colony growth for three months. We found that queen-rearing colonies that were fed treated pollen had a reduced capacity for queen cell production, and we observed a significant decrease in queen survival during development within constructed queen cells. Queens that developed within cell-builder colonies that were fed treated pollen were later found to have reduced sperm health relative to queens reared within control colonies, and once installed in a full sized colony these queens were observed to have reduced brood viability. Our findings suggest that queens which survive development within highly contaminated environments may still be compromised despite having successfully mated. Testing oral and contact exposure mixtures developed from previously recorded in-hive residues affords a new degree of realism in pollinator toxicology, and by tracking the downstream implications of our treatments we can better understand exposure associated fitness costs resulting from developmental chemical environments.

[P.03.46] Pesticides in Pollen: Results from the US National Honey Bee Disease Survey

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The potential impacts of pesticides on honey bee health have been the focus of many recent research efforts. Over seven years from 2011-2017, we surveyed pesticide residues in stored pollen (bee bread), as well as disease incidence as part of our US National Honey Bee Disease Survey. More than four-fifths (81.8%) of all samples (n = 1059) collected had at least one pesticide product detected, with a mean of almost three compounds (2.77) per sample. Overall 5.9% of the samples exceeded a Hazard Quotient (HQ) of 1,000, equivalent to 10% of a honey bee's lethal dose if consumed for the duration of her 10 day nursing phase (LD50), while 0.57% (n = 6) exceeded a HQ of 10,000 equivalent to the LD50. Two-thirds of samples (65.8%) had at least one varroacide residue, and varroacides made up just over 40% of all pesticide detections (n = 1,226 of 2,933). Fungicides were fairly common comprising 22.2% of all pesticide detections (n = 650), with at least one fungicide detected in 29.8% of samples (n = 316), and up to ten different fungicides detected in a single sample. Very few samples (4.8%; n = 51 samples) were contaminated with neonicotinoids. This research is the most comprehensive assessment of real world pesticide exposures in US managed colonies to date. By linking exposure with colony morbidity measures, this study highlights areas of special concern for researchers, policy makers and extension personnel dedicated to improving pollinator health.

[P.03.47] It's complicated: How pesticide applicators understand hazards to honey bees (and inconsistencies in how US pesticide labels present these hazards)

A. Melathopoulos [1], R. Kachadoorian [2], G. Uribe [2], M. Bucy [1]
[1] *Oregon State University, Corvallis, USA,* [2] *Oregon Department of Agriculture, Salem, USA*

The Oregon Department of Agriculture created incentives for licensed pesticide applicators to be trained in how to protect bees from pesticide exposure, following a string of high-profile bee poisonings following the treatment of shade-trees with insecticides in 2013 and 2014. We initiated a new training program in 2016 to determine the extent that applicators could be trained to understand honey bee warnings on pesticide labels. We used electronic clicker technology to administer pre- and post- training evaluations and discovered that: 1) while fewer than a quarter of applicators could interpret the toxicity of a pesticide to honey bees, or residual toxicity statements on pesticide labels, 2) their comprehension increased to over 95% following a 60 minute training. The effectiveness of our training, however, was predicated on pesticide labels communicating acute and residual toxicity using standardized language recommended by the US Environmental Protection Agency (USEPA). To determine the extent to which pesticide labels reflect this standardized language, we compiled a database of 232 insecticide labels reflecting 16 high-risk exposure scenarios to Oregon honey bee colonies. We found that: 1)

31.2% of these labels contained at least one deviations from USEPA's recommended language, 2) these deviations were not limited to any one particular scenario or insecticide subgroup and 3) the most common deviation was between the language used to signal the level of toxicity and laboratory-derived acute toxicity values for honey bees. While our findings underscore the importance of applicator education in reducing pesticide exposure to honey bees, it suggests trainings need to incorporate existing inconsistencies in label language to be more effective.

[P.03.48] Toxicity of synthetic and natural acaricides to worker honey bees (*Apis mellifera*) and their impact on the expression of detoxification-related genes

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In this study, honey bees (*Apis mellifera L.*) were exposed to LD05 and LD50 doses of five commonly used acaricides for controlling the parasitic mite, Varroa destructor. LD50 values at 48 h post treatment showed that tau-fluvalinate was the most toxic, followed by amitraz, coumaphos, thymol and formic acid. However, the hazard ratios, which estimate the hive risk level based on a ratio of a standard dose of acaricide per hive to the LD50 of the acaricide, revealed that tau-fluvalinate was the most hazardous followed by formic acid, amitraz, coumaphos and thymol. The expression of the honey bee acetylcholinesterase gene increased after treatment with the LD05 and LD50 acaricide doses and could distinguish three patterns in the timing and level of increased expression between acaricides: one for amitraz, one for tau-fluvalinate and formic acid, and one for coumaphos and thymol. Conversely, changes in cytochrome P450 gene expression could also be detected in response to all five acaricides, but there were no significant differences between them. Among the acaricides tested, amitraz, coumaphos and thymol appear to be the safest acaricides based on their hazard ratios, and a good marker to detect differences between the effects of sub-lethal doses of acaricides is monitoring changes in acetylcholinesterase gene expression.

[P.03.49] Comparative survival of *Apis mellifera* worker adults and brood exposed to thiamethoxam and fungicides

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Honey bees (*Apis mellifera*) are frequently exposed to combinations of pesticides in pollen and nectar, including the neonicotinoid thiamethoxam and the fungicides prothioconazole and boscalid. Worker honey bees may differ in their susceptibility to chronic pesticide exposure depending on life stage. To compare the sensitivity of adult workers and worker brood to dietary neonicotinoids and fungicides, we chronically exposed newly emerged workers and worker larvae to thiamethoxam and/or prothioconazole or boscalid and monitored survival and hypopharyngeal gland development. We found that adult workers were more sensitive to chronic dietary thiamethoxam compared to worker brood. Co-exposure to fungicides did not enhance effects of thiamethoxam, regardless of life stage. Adult workers chronically exposed to 40 µg/kg thiamethoxam had significantly reduced survival ($P < 0.001$) compared to control; however, worker brood did not experience significant ($P<0.05$) decreases in survival unless exposed to a 23 times greater dose of thiamethoxam (1 mg/L or 909 µg/kg). Hypopharyngeal gland development was not significantly different between pesticide exposed adults and laboratory controls. These results show that (1) adult workers are more sensitive to thiamethoxam compared to worker brood, (2) fungicide co-exposure does not alter the effects of thiamethoxam on worker survival.

[P.03.50] Bee exposure to pesticide cocktails: origin, distribution, and risk

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The honey bee (*Apis mellifera L.*) is an important pollinator that guarantees food safety and biodiversity. Honey bee health has been in decline, and while there are many threats to bee health, pesticide exposure has been considered as a major one. To begin to understand this threat, the actual pesticide residues to which bees are exposed to in the field needs to be measured, from multiple locations and over time. The widespread use of pesticides exposes bees to multiple chemicals simultaneously (chemical mixtures), creating pesticide cocktails that could cause detrimental synergistic side-effects. However, the risk assessment of pesticides is usually limited to individual pesticides. Thus, we examined real world pesticide exposures using multiple bee health monitoring surveys (years: 2011-2017, N = 1,114). We monitored bee colonies that were located across 43 USA states, and others that were specifically used for pollination of USA target crops. Pesticide residue levels were measured in bee-collected corbicula pollen or beebread. We focused on the role played by chemical mixtures, highlighting the most frequent binary pesticide combinations that could cause amplified adverse synergistic effects. Our results show that the majority of samples contained pesticide cocktails, up to 21 chemicals per sample, that the combinations that co-occurred more frequently were clothianidin-atrazine (100%), and pyrimethanil-boscalid (90%), and that a relevant number of combinations could cause synergistic effects. We report the results related to the origin (i.e. target vs. non-target crops), distribution across space (US states,

and specific crops) and time (year, and month), toxicities, and consequent risks for bees caused by field-realistic pesticide combinations. We discuss the results from a bee health and Risk Assessment perspective, and suggest pathways forward for addressing the impact of interactive (i.e. synergistic) effects of multiple pesticide exposure on bee health.

[P.03.51] Chronic exposure effects of sufloxaflor under laboratory and field conditions

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Sufloxaflor is one of the sulfoximines which constitute a highly effective group of pesticides against a wide range of sap-feeding insect. However, it also affects the non target insects such as the honey bee. In order to see if there are chronic exposure effects on the bees, a laboratory and a field experiment was contacted, where sufloxaflor was administered via the food. Sufloxaflor was fed to the bees at a dose of 50ppb in sugar solution or in both sugar solution and pollen patties. For the laboratory experiment, bee survival, rate of mortality, amount of consumed food and the amount of fat body was accessed. For the field experiment, colony population, amount of brood and dead bees in frond of the colonies was accessed. The preliminary results have shown no difference in any variable between treated and control bees in the laboratory but a significant difference was detected between treated and control colonies in terms of amount of brood. Most of treated colonies did not manage to overwinter successfully. Although preliminary, the results demonstrate the adverse sublethal effects of this substance that recently has been introduced to many countries in Europe in order to be used against insects resistant o neonicotinoids. Further experiments are needed to demonstrate the real impact on honey bee colonies and bee's physiology and behaviour.

[P.03.52] Efficacious biological insecticide proven safe for honeybees in semi-field conditions in Quebec (Canada)

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Many insecticides have been proven to be harmful to honey bees in different aspects. Safety for honey bees regarding an efficacious alternative is presented. Twelve test tunnels (experimental and controls) were put up at Le Centre de recherche en sciences animales de Deschambault (CRSAD) in summer 2018. One hive was placed in each tunnel along with one water feeder (10cm x20cm). Dead bee traps were placed on each hive. Colonies were free of disease (e.g. Varrosis, Nosemosis, Amoehiosis, Chalkbrood, Sacbrood, American or European foulbrood) or pests (Varroa destructor). The tunnel surface areas (20mx30m) were covered with Trifolium pretense, T.repens and Brassica spp. Flowers were attractive to honey bees and bloomed during the two exposition periods (21 June to 1 July 20 and 13 August to 23 August 2018). At nightfall, the vegetation inside each of the experimental tunnels was sprayed with BioCeres® WP (Beauveria bassiana ANT-03) at the maximum label recommended application rate (6 g/L) and a volume rate of 200-400 L/ha. The control tunnel vegetation was treated with tap water at the same volume rate. Tunnels and hives were monitored for honey bee and colony health (behaviour, foraging, mortality, brood and food stocks). Honey bees in tunnels of both treatments were foraging, drinking and flying in a similar fashions. For adult bees, the number of specimens collected in the dead bee traps from each treatment were low and not statically different between treatments. On rare occasions, more drones and larvae were collected in dead bee traps of the BioCeres tunnels during hive seclusion. The specimens were sent for further laboratory analyses that confirmed that BioCeres spores were not present on or in these dead bees. Brood and food stock data were also shown to be similar. In the light of this study, BioCeres, containing the active ingredient of conidiospores of the strain B. bassiana ANT-03, at the highest recommended doses did not infect, cause sub lethal behavioural abnormalities, nor kill the honey bees under field conditions

[P.03.53] Sublethal pesticides in pollen reduced brood rearing and increased queen loss

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The European honey bee, *Apis mellifera*, the most important pollinator of agriculture crops in the United States, is regularly exposed to an array of pesticides during pollination services. Pesticide residues in bee bread were analyzed for six consecutive years from apiaries in over 35 states. Two insecticides (chlorpyrifos and fenpropathrin), and two fungicides (chlorothalonil and propiconazole) were frequently detected during the survey. We applied field relevant doses, at a) 10% of LD50 of the two insecticides, or b) 2% of the LD50 for the two fungicides or c) a combination of both to treatment groups over a 30 day period. The contaminants were fed to the treatment groups through pollen patties, and incoming pollen was restricted by a pollen trap installed at the entrance of every colony. Foraging activity was recorded weekly at the hive entrance. All treatment groups sent out a higher proportion of pollen foragers compared to controls, though engaged in less overall foraging activity. Additionally, each treatment group consumed less of the pollen patty than the control group. Brood development was tracked from egg to emergence, before and after treatments. Following treatment, total loss of brood prior to emergence was 11.4% for the control group, 20.4%, 29.3%, and 20.8% for fungicide, insecticide and both contaminant groups respectfully. Brood cannibalization was significantly higher in all treatment groups. Queen loss through supersedure occurred in the treatment groups as well.

[P.03.54] Are all honey bee castes affected equally by exposure to thiamethoxam during late larval development?

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Honey bees provide essential pollination services and a number of products used in many industries (food, pharmaceuticals, dentistry, cosmetics, etc.). Out of the three honey bee castes (workers, drones, queen) workers are the most numerous and are extensively used in toxicity assays. As workers are non-reproductively active, their evaluation alone may overlook potential toxic effects on bee reproduction. However, a number of environmental contaminants were already shown to decrease reproductive potential of the honey bee drones and queens.
The objective of this study was to evaluate if the honey bee castes were affected equally by exposure to thiamethoxam (THI) during late larval exposure.
Five honey bee colonies were manipulated to produce synchronized age of worker, drone and queen brood. Larvae of each caste were exposed to one of 4 experimental treatments 1 day prior to capping (worker – day 8, drone – day 10, queen – day 7 of development from oviposition). The treatments consisted of 4 µl of double distilled water containing 0, 5, 50, or 100ng of THI. Post treatment capping, eclosion, and emergence weights were recorded for all castes. Drone and worker post emergence survival was monitored in the laboratory conditions.
Emergence rate in THI100 group was 26.7% and 84.26% for queens and workers, respectively, but was not affected in drones. The emergence weights were significantly decreased in drones and workers exposed to THI100 and workers exposed toTHI50 but not in queens. No treatment affects were observed on drone and worker survival in laboratory conditions.
The results of our study may suggest that honey bee castes are not equally susceptible to THI during late larval stages.

[P.03.55] No adverse effect of Bt Cry9Ee toxin on honey bee brood and adults reared in vitro, *Apis mellifera* (Hymenoptera: Apidae)

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The effects of Bt Cry9Ee toxin on honey bee, *Apis mellifera L.*, survival, developmental rate, larval weight, pollen consumption, and midgut bacterial diversity were tested in the laboratory. Honey bee larvae and adults were reared in vitro and fed a diet that contained Cry9Ee toxin at 0.01, 0.1, 1, and 10 mg/L. Cry9Ee toxin 0.01, 0.1, and 1 mg/L in diet used in this study may represent a value closer to field relevance and the highest concentration is unlikely to be encountered in the field and thus represent a worst case scenario. The dependent variables were compared for groups of honey bees feeding on treated diet and those feeding on negative control (no addition of a test substance), solvent control (0.01 mM Na_2CO_3), and positive control diet (dimethoate 45 mg/L). Bt Cry9Ee toxin did not affect survival, developmental rate, larval weight or pollen consumption. Furthermore, the midgut bacterial structure and compositions were determined using high-throughput sequencing targeting the V3-V4 regions of the 16S rDNA. All core honey bee intestinal bacterial class such as γ Proteobacteria, Actinobacteria, α Proteobacteria, Bacilli, β Proteobacteria, and Bacteroidia were detected, and no significant changes were found in the species diversity and richness between Cry9Ee treatments and laboratory control. The study confirms that Cry9Ee toxin has no adverse effect on honey bee brood and adults reared in vitro.

[P.03.56] Neonicotinoids decrease sucrose responsiveness of honey bees at first contact

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Neonicotinoid insecticides are known to have harmful effects on the behaviour and physiology of many insects. Through pollination services, honey bees are exposed to these insecticides in pollen and nectar. Impaired navigation and decreased foraging activity are some of the negative effects reported for neonicotinoids. We exposed caged foragers to sub lethal acute doses of three neonicotinoids (clothianidin, imidacloprid, and thiamethoxam) and tested them individually for sucrose responsiveness. We also tested the effect of a range of sucrose solutions laced with neonicotinoids on bees previously unexposed to neonicotinoids, to mimic the situation where foragers would first encounter poisoned nectars varying in sugar concentrations. Thus, bees were exposed to the insecticides either in the diet fed to caged foragers for 24 hours before testing or in the test solutions used to measure sucrose responsiveness, or both. We report a detrimental effect on honey bee responses to mid-to-high sucrose concentrations under all experimental conditions. Previously unexposed bees displayed unexpectedly low responses to the higher sucrose concentrations tested. This attenuation of sucrose response is further evidence that neonicotinoids are multisensory disruptors, with potent actions at first contact, against pollinators and other beneficial insects.

[P.03.57] Toxicity to honey bees *Apis mellifera adansonii* of three insecticides used in cotton cultivation in Benin

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Protection and preservation of bees are a pledge in a sustainable and environment-friendly agriculture. The economic contribution of the insect's pollinators to world agriculture is estimated at billions of dollars. However, non-judicious choice of pesticides and absence of good plant protection practices, represent a real danger to bees. The objective of this study was to assess the toxicity of three insecticides commonly used in cotton protection on bees *Apis mellifera adansonii* in Benin. In this context, bees were taken from hives and transported to the laboratory. Active ingredients and different doses used were as followed: Emamectin benzoate, beta-cyfluthrin+imidacloprid and lamdex+chlorpyrifos at doses of: 2150 nanograms per bee, 125 nanograms per bee, 75, 50, 25, 12.5×103, 6, 2.5 and 1.25-0.96 ng per bee, depending on the active ingredient. Each treatment included three replications consisting of 25 bees. Bees were anaesthetized with ether before treatment. Each bee received by topical application on the pronotum, 1 µL of the formulation. Observations were made after 2, 10, 24 and 48 h. Results indicated that, even the lowest doses of the pesticides: 0.96, 7.25 and 21.5 ng per bee showed mortality higher than 90%, 48 h after application. An insecticide with active ingredients including beta-cyfluthrin (45 g L-1), which is a pyrethroid and Imidacloprid (100 g L-1), a neonicotinoid actually forbidden in many European countries showed very high toxicity to honey bees, indicating values of the LD50 varying from 19.9 ng per bee for 10 h exposure to 1.1×10-2 ng per bee for 18 h and 5×10-4 ng per bee for 36 h. Results of the study indicated the urgent need of good plant protection practices in the frame of sustainable agriculture and bee's preservation and conservation. Promotion of strategies as part of the dissemination of good agricultural practices in plant protection is a guarantee which should ensure sustainable agriculture and environmental, human and biodiversity protection. These studies showed the urgency of the integrated plant protection in order to promote good agricultural practices for honey bee's protection.

[P.03.58] Transcriptome Analysis of Newly Emerged Honeybees Exposure to Sublethal Carbendazim During Larval Stage

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There are increasing concerns regarding the impact of agrochemical pesticides on non-target organisms. Pesticides could cause honeybee abnormal development in response to neurotoxins such as neonicotinoid. However, knowledge of carbendazim, a widespread fungicide in beekeeping practice, influencing on honeybee (*Apis mellifera L.*) brain development is lacking. Large-scale transcriptome approaches were applied to determine the changes in global gene expression in the brains of newly emerged honeybees after carbendazim exposure during the larval stage. To further understand the effects of carbendazim on the brain development of honeybees, the functions of differentially expressed genes were compared between the treatment and control groups. We found that neuroregulatory genes were down-regulated after carbendazim exposure, which suggest the neurotoxic effects of this fungicide on honeybee nervous system. Carbendazim exposure also altered the expression of genes implicated in metabolism, transport, sensor, and hormone. Notably, larvae in the carbendazim-treated group observed longer time to shift into the dormant pupal state than the control group. Moreover, a low juvenile hormone and high ecdysone titers were found in the treatment group compared to control group. The data is the first report of neurotoxic effects on honeybee caused by carbendazim, and the sublethal carbendazim may disturb honeybee development and is a potential chemical threating the honeybee colonies.

[P.03.59] Cypermethrin alter the midgut morphology of *Tetragonisca angustula* (Latreille, 1811) stingless bee

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Stingless bees are important pollinators of crops and natural environments. The expansion of agricultural activities and the extensive use of agrochemicals to control pests can also be harmful and toxic to non-target insects such as bees. *Tetragonisca angustula* species is a floral visitant of several crops in which the cypermethrin is being used to control pests. The foragers bee were collected from commercial hives on the campus of the State University of Maringa, Parana, Brazil. The survivor worker bees exposed to a diet containing doses of cypermethrin Nortox 250 EC at 1.031 × 10-6 g a.i./mL, 4.125 × 10-6 g a.i./mL and 8.250×10-6 g a.i./mL for 24 and 48 h were dissected and the midgut was processed for light and scanning electron microscopy. Individuals exposed for 24 and 48 h had morphological alterations in the digestive and regenerative cells of the midgut when compared to the control bees. We detected the absence of regenerative cells in the nests for all concentrations tested. After 24 h of exposure at the lowest concentration 1.031 × 10-6 g a.i./mL, the midgut of T. angustula workers showed basal lamina epithelium detached and increased apocrine secretion. In addition, prismatic digestive cells became cuboid and disorganized digestive cells were eliminated to the lumen. The alterations were intensified at higher concentrations as cytoplasm vacuolization and disruption of longitudinal muscular fibers at 4.125 × 10-6 g a.i./mL. We also identified disorganization of the peritrophic membrane and high secretion of digestive cells in the midgut of bees by intake the insecticide at 8.250 × 10-6 g a.i./mL in the diet. There were pronounced alterations after exposure for 48 h until the whole destruction of the intestinal epithelium at 8.250 × 10-6 g a.i./mL

concentration. For all doses and periods tested occurred the loosening, disorganization and dilation of the longitudinal muscle of midgut. However, at the highest concentration for 48 h the longitudinal fibers suffered a narrowing and circular muscles almost disappeared. Thus, several alterations in the midgut cells suggest that the toxicity of cypermethrin compromises the physiology of T. angustula stingless bee species.

[P.03.60] Interactions between pesticide diomethoat and pathogen nosema in honey bees

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In the recent decades pollinating insects, especially bees have declined in species richness, geographical range and abundance. There are several factors affecting the health and survival of managed bee colonies such as the spread of parasites and pathogens, changing cultural and commercial beekeeping practices, agricultural and apicultural pesticides both in the field and in the hive. The most significant threats to managed bee colonies are increasing the prevalence of parasites and pathogens as well as excessive use of pesticides in agriculture. Changes in the physiology of pollinators have been observed when exposed to pesticides. Pesticides affect pollinators immunity systems and physiology, which in turn makes them more susceptible to various pathogens.
The aim of the research is to find out how does the pesticide dimethoate affect the metabolic rate of honey bees that are already infected with Nosema apis, N. ceranae or the mix of the two.
For the research, 12 hives which were separated into isolated pavilions, were used in spring 2018, with 6 frames and a young italian race queen in each hive. The colonies were fed with infected sucrose syrup (5mil spores per colony) either of the pathogens or the mix of both chronically during three weeks. Honey bee individual mortality was observed by creating 9 mini-colonies (20 worker bees) per each treatment group and feeding them with sugar syrup mixed with dimethoate (20 µg kg-1). For metabolic rate measurements (rate of CO2) the extra bio-assays with similar treatments were created. Flow-through respirometer LI-7000 (LiCor, Lincoln, Nebraska, USA) was used for this purpose. A flow-cytometer was used to determine the presence of the disease.
The results showed that none of the treatments for the bees had a plausible effect on the metabolic rate of honey bees. However, the individual mortality increased significantly after treatment with the mix of the two pathogens or with N. ceranae. The N. apis did not increase the mortality compared to control bees, however, at the day 16 after treatment started, we saw an increase in mortality of N. apis treated honey bees.

[P.03.61] Distribution of Carbon Microparticles in Honey Bee Colonies for Potential Pesticide Protection

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Honey bees (*Apis mellifera*) are the most commonly used species to provide pollination service in agriculture because of the ease of transportation and high abundance during crop bloom. However, pesticides widely used in farmlands to control or eliminate pests pose a potential risk to honey bees. Because pesticides have shown various negative effects on honey bees such as learning ability, orientation, brood care, promoting Nosema infection and reproduction. Various methods have been employed to remove pesticides in other systems (eg. soil and aqueous solutions) and one such method is the use of activated carbon. Based on similar chemistries, a form of carbon microparticles (CM) was developed to mix into honey bee feed with the intention of absorbing pesticide compounds, rendering them nonfunctional, before they harm the bee or the colony. However, there needs to be an understanding of; where or if bees store CM in hive products or feed it to larva in colonies after they consume CM added to bee feed. The purpose of this research is to determine the distribution of CM in full-size honey bee colonies. Colonies were fed CM in sugar syrup and be monitored regularly. To detect CM in colonies and hive products, samples were collected from nectar, pollen, wax, young larvae, and old larvae during the experiment. The CM in samples was quantified using thermogravimetric analysis. CM was found in stored nectar and pollen samples and implications of these findings will be discussed. It is likely that methods and findings from this research could be applied to understand the use and distribution of other honey bee feed additives within the hive.

[P.03.62] Mass death and collapse of bee colonies in Brazil, surveyed by Bee Alert application, 2013-17

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Two major problems in today beekeeping are colony loss and CCD (Colony Collapse Disorder) of bees, which are causing the world population decline of many species. In March 2014, an online survey was launched in order to assess the evolution of bee colony losses and CCD events in Brazil. Occurrences happened from January 1st, 2013 to December 31st, 2017 were accepted to be posted in a web survey hosted in www.nobeenofood.com/beealert, as well as in apps for smartphones and tablets, from interested collaborators in making their registrations. Three hundred twenty two qualified and validated participants completed the questionnaires and were accepted to compose the Bee Alert application database in that period. During visits to these apiaries, 114 live and dead honeybee samples were collected for analyses of pesticide residues by means of mass spectrometry (UHPLC-MS/MS) and QuEChERS method. Statistical analysis showed that pesticides are reported as the main cause of colony loss and CCD in the country. Total colony loss calculated for the period (2013-17) was 49.7% with (95% CI 46.3-53.2%), and average colony loss was 64.7% with (95% CI 61.0-68.4%). Neonicotinoids and fipronil lead pesticide lists (55.9%), and São Paulo state holds 45.7% of honeybee casualties in Brazil. Toxicological analyses in all samples showed multiple contamination with higher indices by fipronil with frequency of 55.3% and amplitude of (0.7-23.540 ng/g), thiamethoxam 20.2% and (0.6-13.6 ng/g), imidacloprid 3.5% and (4.5-16.2 ng/g), nitenpyram 1.8% and (3.8-7.4 ng/g) and thiacloprid 0.9% (1.6 ng/g). Acetamiprid, clothianidin and dinotefuran residues were not detected (<LOD), possibly due to degradation and metabolization. Neonicotinoids and fipronil residues had higher frequencies and amplitude in honeybees collected near sugarcane plantations and orange orchards in northwest São Paulo and other agro-industrial rural landscapes. Africanized honeybee (*Apis mellifera*) and the native bee jataí (*Tetragonisca angustula*) were the most harmed species. These systemic pesticides are the main mechanisms of mass death and collapse of bee colonies in Brazil, according to this research.

[P.03.63] Sublethal effect of pesticides exposure at the larvae stage on honey bee queens

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The domestic bee (*Apis mellifera*) is the most economically important pollinator in the world. Honey bee pollination contributes to the biodiversity and agriculture productivity. Since several years the decline in honey bee populations worldwide is alarming. Colony mortality can be explained by multifactor effects among which the expansion of pesticides. Recently, several methods and technics were developed to evaluate the pesticides' effect on the mortality of honey bee but also of the effect of sub-lethal doses on their life history parameters, neurobiology and behavior. Previous studies were mainly focused on the workers. Here we will focus on honey bee queen as little is known about the sublethal effect of pesticides on their physiology and reproductive abilities.
An important tool for the research on pesticides is the rearing of honey bee larvae in vitro to control chronic pesticide exposure on larvae development. However, such technic is no applicable to honey bee queens. We developed a method for the rearing of queen larvae allowing exposure to pesticide during queen larvae development. We will present the rate of queen rearing success. Emerging queens will be observed until mating and sacrificed few days later to compare their ovarian development and their pheromone composition of the mandibular gland. The results are currently under analysis. The consequences of pesticides will be discussed.

[P.03.64] Do Beauveria bassiana and Metarhizium anisopliae affect the workers' survival and the rearing of Africanized *Apis mellifera L.*, 1758 (Hymenoptera: Apidae) queens?

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Apis mellifera are disappearing due to the Colony Collapse Disorder (CCD) and the exacerbated and incorrect synthetic chemical insecticides use is listed as one factor. Unlike this, the use of biological control of pest insect with entomopathogenic fungi is considered security, however, the effect of these on *A. mellifera*is still few elucidated. In this sense, the objective was to evaluate the effect of *Beauveria bassiana* (IBCB 66) and *Metarhizium anisopliae* (IBCB 425) on the workers' survival and the rearing of Africanized *A. mellifera* queens. In the laboratory, four bioassays were performed with the entomopathogenic fungi (1×108 conidia.ml-1): 1) contact of workers on a sprayed glass surface, 2) contact of workers on treated eucalyptus leaves, 3) spraying treatment on workers and 4) ingestion of candy paste incorporated with the treatments. Sterilized distilled water was used as a control. Each treatment consisted of 100 honey bees, arranged in five replicates. After the bioassays were carried out, the workers were transferred in groups of 20 individuals to PVC cages, covered with voile, containing a piece of cotton soaked in water and a portion of candy paste (27 ± 2 ° C, RH 70 ± 10% and 12/12h L/D). The workers' longevity was evaluated from one to 240 hours. Two cycles were performed to queen rearing:1) *B. bassiana* and sterilized distilled water and 2) *M. anisopliae* and sterilized distilled water. For this, the transfer of worker larvae to acrylic cells containing royal jelly incorporated with the treatments was performed, with the emergence of the queens was monitored and morphometric analyses performed. *Metarhizium anisopliae* caused a reduction in the workers' longevity in the four bioassays performed (73.4, 117.7, 139.9 and 126.9 hours, respectively) and *B. bassiana* caused a reduction in the longevity of the workers only in the direct spray bioassay (159.4 hours) when compared to workers from the respective control. In the field, the entomopathogenic fungi did not affect the production and morphometry of the *A. mellifera* queens produced and their time of emergency. Despite this, *B. bassiana* and *M. anisopliae* are considered safe to produce *A. mellifera* queens in the field.

[P.03.65] An efficient approach to monitor the residues level of neonicotinoids: a study case in Romania

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In the last year numerous researches were focused on the impact of neonicotinoids on honeybees and other pollinators. Honeybees are an excellent biosensor as, by their biology and their management can be used to collect valuable samples which offer important information about the environmental contamination. Our researches, carried out in 2018 in Romania, aimed to establish the level of neonicotinoids level in bee products, honeybees and targeted plants. The studied cultures were rape, sunflower and corn experimental fields, located in different areas of Romania. The evolution of honey flows intensity correlated with natural conditions (temperature and humidity) were monitored by electronic hives (Simbee). The samples from different matrices were collected and prepared by specific methods according to the requirements of the accredited laboratories. A series of peculiarities regarding the accuracy of sample collection and preparation methods were noted and will be presented together with the obtained results regarding the level of neonicotinoids residues in different matrices and the impact on honeybees.

[P.03.66] Bees or no bees, this is the question: Deaths of bees and CCD (Colony Collapse Disorder)

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The “Bee Or Not To Be” NGO of Ribeirão Preto - SP, Brazil is an association which main purpose is to defend the bees and protect the environment. Among the more important actions of the NGO we highlight: 1- the creation of the Bee Alert application that allows beekeepers, stingless beekeepers and researchers to register online on a world map, bee occurrence, death of bees and CCD (Colony Collapse Disorder); 2- an Environmental Educational Project “No Bee No Food: Notebook of Activities for Environmental Education” direct toward children from 8 to 10 years old teaching basic bee biology and pollination with surprising results and 3: a Ph.D. thesis by one member of the NGO, Dr. Dayson Castilhos of UFERSA, Mossoró - RN through the usage of the Bee Alert application database and which carried out toxicological chemical analysis of live and dead bees samples contaminated with pesticides after aerial spraying in agricultural landscapes in Brazil. Bee Alert records showed during the first four–year survey (2013-2016) that an estimated 770 million bees were killed, mainly caused by the indiscriminate use of neonicotinoid and Fipronil pesticides. Higher frequencies of deaths occurred in the states of SP and RS. The most affected bees were the Africanized bees (*Apis mellifera L.*) followed by some native stingless bees. Brazilian beekeeping has suffered in the last three years an irreparable loss of bees in several states; more frequently in the states of São Paulo and Rio Grande do Sul, where pesticides are cited as the main cause of bee deaths and CCD cases. In those states many beekeepers are giving up beekeeping, due to high colony losses recorded, and lack of support in reimbursing the financial losses. More than 6.000 colonies of Africanized honeybees were killed only in Piaui state, Brazil, in last four months. The question that arises is: Bees or No Bees: Is it possible for bees to resist the indiscriminate use of pesticides in the world?

[P.03.67] Challenges of organic beekeeping in monoculture farms; Based on researches performed in California, Washington State, Vancouver, and Iran (North, West and South)

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The purpose of this research was to examine the prevailing challenges that beekeepers encounter with organic beekeeping in monoculture farms, as well as to investigate the economic and practical aspects of it.
This research was done on commercial beekeeping operations during and after pollination season, in 6 locations, in 3 countries, California, Washington State, Vancouver, and Iran (North, West and South). Honey and Propolis were tested against the presence of 37 common herbicides; no residues were found. However, a significant loss of bee colonies was experienced due to malnutrition and diseases caused by the lack of pollen diversity. The bee colonies were infected with a high level of EFB, Carana, and Nosema Apis, and had symptoms of diarrhea (mostly in California, Washington state and Vancouver).
If beekeepers have to place their hives in monoculture farms, as it is the case in commercial beekeeping operations, a sufficient amount of nutrition should be provided to the bees, to prevent malnutrition and susceptibility to diseases. It is also important to be aware of Varroa mites and epidemic diseases that might spread from other infested and contaminated apiaries.
In conclusion, organic beekeeping in monoculture farms is neither practical nor economic, due to the high risks of contamination and infestation, as well as the costs involved in running the operation. In this presentation, all these key factors will be discussed in further details.

[P.03.68] Assessment on the effects of Agro-chemical Applications on Honeybee production in Selected zones of Tigray Region, Northern Ethiopia

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Assessment on the effects of agrochemical applications on honeybee production was conducted in eastern; south-east and central zones of Tigray region from September, 2014 up to June, 2015 to assess the types of agro-chemicals and their effects on honeybees and their products. Questionnaire survey and observation methods were used for the study. In the questionnaire survey, 384 beekeepers (350 male and 34 female) were interviewed. From the total of 384 beekeepers interviewed, 52.3% of them are recognized as pesticides users. Moreover, there was significant variation in use of agrochemicals among beekeepers in the study districts ($p<0.01$). Agrochemicals were used for the purpose of pest control (98%), weed control (84.6%), for veterinary use (12.4%) and malaria transmitting anopheles mosquito

repellent (3%). According to the respondents, the most used brands of agro-chemicals were, Agro- 2-4-D (85.6%), Malathione (73.7%), Karate (39%), Dimothoate (33%), Ridomil (28.9%), Mancozeb (27.8%), Dursban (24.5%),Fenithrothion (24.5%) and Diazinon (22.8%). Majority of the respondents apply the chemicals during the morning time (48.5%) followed by day time (21.5%), evening (17.5%) and at any convenient time throughout the day (12.5%). The respondents claimed that within the last four years, 219, 219 and 34 honeybee colonies were recorded as absconded, dwindled and died due to indiscriminate application of aforementioned chemicals in the study districts, respectively. Therefore, there should be strong communication between beekeepers and crop growers while spraying. It is important to advise people in selecting and applying less hazardous chemicals to honey bees before blooming and when honey bees are not foraging in the field.

[P.03.69] French Beekeepers against neonicotinoids: a 20-years fight

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In 1995 in France, there were 85,000 beekeepers and 1.4 million hives which produced 33,000 tonnes of honey. Ten years later, in 2004, the sector had lost 15,000 beekeepers, the hive number had dropped to 200,000 units, and honey production was reduced by one third, thus falling to 22,000 tonnes. Between these two dates, the neonicotinoids appeared in the French fields causing massive depopulations of hives, more than 40% of winter mortality in some areas and important harvesting decreases.
From the beginning, producers of these new insecticides contradicted their effects on bees, despite increasingly sophisticated scientific studies demonstrating it, and in particular the toxicity of these molecules at doses having been found in nectar and pollen. The scientific battle was starting.
The government also acted with a high inertia. For example, European regulations risk quotient was not applied. The French state was also reluctant to give public information to beekeepers.
Given this inertia and given the remained mortalities, French beekeepers decided to resolutely involve themselves in a fight at the media and legal scope. In several years, the legal battle has achieved many victories in the civil and administrative courts. All this was widely publicized and has led in the 2000s to the withdrawal from the market of the Gaucho on sunflower and corn (Imidacloprid) and of the Regent (Fipronil).
Recently, in 2016, thanks to the support of committed members of parliament, a French law (the Biodiversity law) banned all agricultural uses of neonicotinoid insecticides (imidacloprid, thiamethoxam, clothianidin, acetamiprid and thiacloprid). This French regulation goes further than the European ban. This victory is the result of 16 months of advocacy, coordinated with a dozen environmental NGOs and agricultural organizations. The ban came into effect in September 2018.
In 2017, following the French authorization of the new-generation neonicotinoid sulfoxaflor, beekeepers again mobilized and obtained in 2018 the legal prohibition of sulfoxaflor.

[P.03.70] Assessing exposure and sublethal effects of pesticide mixtures for ground-nesting bees

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In agricultural environments, wild bees can be exposed to pesticides through various routes, including oral exposure via ingestion of contaminated pollen and nectar, and contact exposure with plant materials, dust, and sprays. The effects of a growing number of pesticides on bees through these routes of exposure have been heavily studied in recent years. However, pesticide exposure may also occur via nesting sites and nesting materials, such as leaves, wood or mud. For instance, bee species that nest or hibernate below ground may come into contact with pesticide residues in the surrounding soil. This additional exposure route could be extremely important for both ground-nesting bees and bumble bee queens but has yet not been considered. Bees foraging in agroecosystems are also exposed to pesticide mixtures since farmers frequently use multiple pesticides for managing different pests and diseases in crops. While most pesticide risk assessments in pollinators have focused on insecticides as an obvious threat to bees, it is now apparent that fungicides have the potential to enhance the toxicity of insecticides. Yet, the combined toxicity of both agrochemicals in wild bees remains understudied. Using the hoary squash bee (Peponapis pruinosa) and the common eastern bumble bee (Bombus impatiens) as agriculturally-relevant model species, our current research aims to: 1) explore soil as a route of pesticide exposure for bees that nest or hibernate underground, and 2) address the potential sublethal effects of a combined exposure to field-realistic levels of fungicides and insecticides on the development of squash bees, and the hibernation success and colony initiation of bumble bee queens. We used a combination of field and semi-field methods to assess the extent of translocation of pesticides from soil to ground-nesting bees and to generate field exposure estimates of overwintering bumble bee queens to pesticide residues in soil. Considering that the vast majority of bees around the world are ground nesters, it is urgent to understand how these important pollinators could be impacted by pesticide mixtures through various routes of exposure.

[P.03.71] Evaluation of productive and sanitary parameters in honey bee colonies (*Apis mellifera*) after of the exposed with carbaril, in chihuahua, Mexico

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The exposure of bees to pesticides in crops has harmful effects on the health of the colonies, causing immediate death in most cases, there are few studies regarding the residual effects of these substances in the colonies. The objective of the study was to determine the residual effect of the pesticide Carbaryl (Carbamate) on the productive parameters of colonies that were exposed, which is a pesticide commonly used in the cultivation of apple trees. The work was carried out in an apiary located in the municipality of Chihuahua, Mexico, in the desert area during the harvest period of mesquite honey in the month of May and 30 days after a process of pollination of apple trees. For the experiment, used 60 colonies of *Apis mellifera* in production stage were divided in three groups of 20 beehives: Group 1: Colonies that did not participate in the process of pollination of the apple tree, Group 2: Colonies that participated in pollination of orchards conventional apple (exposed to chemical pesticides) and Group 3: Colonies that participated in pollination of organic apple orchards (Not exposed to chemical pesticides). The variables analyzed were size of population in the hive, size of nest, quality of egg laying, quantity of food reserves, production of honey and health status of the colonies (mortality and presence of other pathologies). According to the parameters evaluated, it was determined if the colonies had suitable conditions for production. At the end of the experiment, it was observed that the colonies of the group exposed to chemical pesticides showed lower performance in the parameters evaluated (40% of suitable colonies) compared with the other two groups: without participation in pollination (90% of suitable colonies)) and pollination of organic orchards (70% of suitable colonies), also low productivity and bees death was observed 30 days after the exposure to pesticides. This reveals that substances such as carbaryl can have residual effects on the health and productivity of colonies chronically.

[P.03.72] Does genetically modified maize pollen affect the longevity of Africanized *Apis mellifera L.* (Hymenoptera: Apidae)?

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Apis mellifera is relevant in the pollination process, however, bee's population is decreasing due to colony collapse disorder (CCD). CCD may be related to many factors, but there are few studies about the influence of genetically modified organisms (GMO). Thus, the objective was to evaluate the effects of GM maize pollen on the longevity of Africanized *A. mellifera*. Newly emerged workers were used (0 to 24 hours) and the pollen (PM) was collected from three GM maize cultivars that express Bacillus thuringiensis (Bt) protein (PM1-Cry1Ab; PM2-Cry1F, Cry1A.105, Cry2Ab2 and, PM3-Cry1F, Cry1A.105, Cry2Ab2) as well as pollen from a conventional maize cultivar. Also, a treatment without pollen was used for each experiment. The pollen was administered in two types of feed, 1) mixed in a liquid diet and in 2) a Candy paste, for each 1 mL of liquid diet and 1 gram of solid diet was added 20mg of pollen. After observing the consumption of all the diet initially has been given, a diet without treatments was provided. The honey bees were stored in PVC cages and kept in climatized room (27 ± 2°C, 60 ± 10 U.R). Each treatment consisted of five replicates, with 20 honey bees per replicate. The evaluation was performed from one to 240 hours, counting the number of dead honey bees. It was verified that in the liquid diet test, pollen from Bt corn with Cry1Ab (PM1) protein was the only one that reduced the longevity of the workers of *A. mellifera* to 85.85 hours, while the workers fed with the non-Bt pollen average longevity of 173.74 hours. The workers fed with pollen from the cultivars PM2 and PM3, had mean longevity of 132.19 and 160.30 hours, respectively. It is noteworthy that both feature PowerCore technology with Cry1F, Cry1A.105, and Cry2Ab2 proteins. In the solid diet, there was no reduction in the longevity of *A. mellifera* workers fed with different types of pollen. Thus, it can be inferred that, according to these tests, the ingest of Bt pollen by Africanized *A. mellifera* workers in the short term does not interfere in their longevity.

[P.03.73] Application Method Matters: Pesticide Residues in Pollen and Nectar Vary Depending on the Application Method and Timing

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Oxamyl (Vydate™) is a systemic carbamate insecticide and nematicide that is classified in the US as acutely toxic to bees. This inherent toxicity can potentially be mitigated by using application methods that reduce the potential for bee exposure. A study conducted in crookneck squash (Cucurbita pepo) measured residues of oxamyl in pollen and nectar after three separate application scenarios: broadcast application immediately before crop planting, drip chemigation treatment during early flowering, and foliar application during early flowering. Pollen and nectar were sampled directly from flowers, from foraging bees and from pollen traps 15 times during the course of flowering. Oxamyl residues in pollen and nectar were highest following the foliar application during flowering, lower following the drip chemigation treatment during flowering, and lowest following the pre-plant broadcast application. Following all application scenarios, oxamyl residues in pollen and nectar declined over the period of flowering, with residues peaking by 10 days after the start of flowering. These data support the conclusion that bee exposure to acutely toxic pesticides can be effectively managed through the use of lower risk application scenarios.

[P.03.74] Behavioral evaluation and biochemical parameters of *Apis mellifera* bees exposed to glyphosate

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The objective of this research was to verify the occurrence of behavioral and biochemical changes in *Apis mellifera* bees exposed to glyphosate by ingestion. The research was conducted at the Laboratory of Genetics and Breeding of Bees in the Department of Animal Science at the Federal Rural University of Pernambuco (DZ/UFRPE). For the bioassay installation, each experimental unit was composed by a PVC container of 15 cm in diameter by 10 cm in height, with 45 bees harvested from the experimental apiary at the DZ/UFRPE. The experiments were carried out in triplicate, with bees kept in an air conditioned environment with an average temperature of 26.3±0.1°C, arranged in three treatments, with 15 bees each, forming the following groups: T1 (control): 1:1 sucrose and distilled water, T2: 360µL glyphosate added to 45ml syrup (1:1 sucrose and distilled water), and T3: 720µL glyphosate added to 45ml syrup (1:1 sucrose and distilled water). The bees were exposed to the treatments for ingestion 1 hour after harvested and the behavioral data were recorded every 30 minutes. The exposure time was 6 hours. For the biochemical profile analysis, the Bradford method was used for total proteins and Van Handel for total sugars and glycogen. The bees were macerated and 1.0mL of 0.1M Sodium Phosphate Buffer solution (pH 7.2) was added to the homogenate. The data were analyzed by the Tukey test ($P<0.05$). The results showed disorientation behavior after 2 hours, T2 (4.53±4.25) and T3 (3.46±4.53), and lethargic behavior after 2h30min, T2 (5.53±5.07) and T3 (6.46±6.38), showing statistically significant differences when compared to the control group, in which the bees remained active throughout the experiment. After 5 and a half hours of ingestion of the solution offered, a mortality rate of 3.3% (n=1) was observed in the T3 treatment. The data for biochemical analyzes did not exhibit significant differences between the treatments. Ingestion of glyphosate, at the concentrations and the time evaluated, caused behavioral changes, but did not induce changes to the biochemical parameters of *Apis mellifera* bees.

[P.03.75] Bio-insecticide azadirachtin promote morphological modifications in the midgut of the stingless bee *Tetragonisca angustula* latreille 1811 (Hymenoptera, Meliponini)

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The excessive use of agrochemicals, specifically insecticides, has been causing concern among honey beekeepers in Brazil, since the disappearance and behavior change of bees may be associated with this exaggerated use of these compounds. An alternative that has been considered less toxic to beneficial insects is azadirachtin. This chemical belongs to the limonoid group, a highly oxidized tetranortriterpenoid. Azadirachtin is a secondary metabolite present in the seeds of the neem (Azadirachta indica A. Juss.) Family Meliaceae, has been used as a bio-insecticide especially in Brazilian organic crops. The goal of this study was to detect the occurrence of morphological alterations in the midgut of *Tetragonisca angustula* after contamination by ingestion with azadirachtin (AzaMax®) diluted according to the package insert for Strawberry crop (3.0 g ai /100 ml). The sublethal concentrations used in ingestion exposure bioassays were 2.25 x 10-3 g ai / ml, 5.25 x 10-3 g ai / ml and 1.12 x 10-3 g ai / ml in the 24 and 48 h periods, with three replicates each. After each period of contamination, bees were sacrificed, dissected, had the midgut removed, were prepared and analyzed by scanning electron microscopy (SEM). The results indicated alterations in the longitudinal and circular muscles of the midgut. Changes such as deformations, loosening and disruption of the longitudinal musculature were observed, presenting greater damages in 24 and 48h, compared to the control. Therefore, it is concluded that, despite not causing mortality, the insecticide AzaMax®, resulted in alterations in the midgut of T. Angustula, which may compromise the viability, behavior and capacity of collecting this species of stingless bees. We suggest that the use of this bio-insecticide be carried out with greater caution and that new studies be carried out to verify possible damages to the meliponiculture.

[P.03.76] Exposure to pesticide residues in wax have invisible effect on honeybee queens (*Apis mellifera*)

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Honeybee (*Apis mellifera*) queen is the most important member of the colony. Despite the fact that several pesticide residues have been found from bee products, the nurse bees are producing pure royal jelly for queen feed. Still, developing larval honeybee queens may be exposed to pesticides via contaminated wax of queen cell cups. Different lipophilic pesticide residues from agriculture as well as apicultrue have been found from honeybee wax (Chauzat & Faucon 2007; Ravoet et al. 2015). The aim of this study was to investigate whether the field realistic concentrations of lipophilic pesticides (tau-fluvalinate and tebuconazole) mixed into the wax are affecting honeybee queen development and maturation.

In two consecutive years (2017 and 2018) field realistic concentrations of tau-fluvalinate and tebuconazole and their mixture were added into organic wax and then queen cell cups were performed. One day old honeybee worker larvae were grafted into treated and untreated queen cell cups and placed into queenless colonies. The parameters measured were: grafted larvae acceptance; queen hatching; newly hatched queen weight; mating success.

Results revealed that the used small pesticide concentrations in wax had significant effect only on certain parameters measured.

Tebuconazole decreased queen acceptance by nurse bees only in one experimental year. However, two different tau-fluvalinate concentrations used in either of the years increased queen weight significantly but no synergy with fungicide was observed. This study helps to fill the gap in knowledge how the sublethal pesticide concentrations can affect honeybee queens even though they are pretty well protected from xenobiotic compounds.

[P.03.77] The effects of a new systemic pesticide, flupyradifurone (Sivanto®) on honey bees: the role of synergism, season, and bee age

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Honey bees (*Apis mellifera L.*) are important agricultural pollinators and are used to model pesticide effects on insect pollinators. Bees can encounter multiple pesticides, leading to potential synergistic interactions. Flupyradifurone (FPF) is a relatively new butenolide insecticide that is systemic and binds to nicotinic acetylcholine receptors. We tested the lethal and sublethal effects of FPF among different worker types and over different seasons and the interaction of FPF with propiconazole, a common SBI fungicide. We report the first demonstration of synergistic effects impairing bee survival (Model Deviation Ratio, MDRMax = 4, Risk Ratio, RRMax = 11; Risk Difference, RDMax = 64) and behaviour (poor coordination, hyperactivity, apathy, RRMax = 15, RDMax = 63) at FPF field-realistic doses. The effects were significantly influenced by worker type and season. Foragers were 4-fold more susceptible to the pesticides than in-hive bees. Both types of worker were more strongly affected by FPF in summer than in spring. Given that risk assessment requires relatively limited tests, only marginally examines bee behaviour and do not consider the roles of season and bee age, these results increase concerns about the safety of approved pesticides, including FPF. Pesticide risk assessment should also test for adverse synergies that may commonly arise upon behaviour and survival.

[P.03.78] Effects of field-realistic concentrations of carbendazim on survival and physiology in forager honey bees (Hymenoptera: Apidae)

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Carbendazim is nowadays widely used to control fungus in various nectariferous crops. Little is known about how honey bees, *Apis mellifera L.* (Hymenoptera: Apidae), respond to carbendazim exposure. In this study, the effects of field-realistic concentrations of carbendazim (4.516, 0.4516, and 0.04516 ppm) on the survival, biomarker enzyme activity (AChE, GST, CarE, and P450), and four antimicrobial peptide gene expression (hymenoptaecin, defensin, apidaecin, and abaecin) in forager honey bees were evaluated. The forager bees were fed with the pesticides for 10 d. The results showed that the field-realistic concentrations of carbendazim did not affect survival; activities of AChE, GST, and CarE; and expression levels of defensin and abaecin in forager bees. However, 4.516, 0.4516, and 0.04516 ppm of carbendazim all significantly inhibited the expression of hymenoptaecin and apidaecin ($P < 0.01$), while P450 (7-ethoxycoumarin-O-deethylase) activity was downregulated by 4.516 ppm of carbendazim ($P < 0.05$). Our results indicate that the field-realistic concentrations of carbendazim may alter the immune response and P450-mediated detoxification of honey bees. Thus, carbendazim should be discreetly used on nectariferous crops during anthesis.

[P.03.79] Chronic high dose thiamethoxam exposure decreases overwinter survival of *Apis mellifera L.*

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Since 2006, North American beekeepers have experienced unsustainable increases in overwinter honey bee colony mortality. Overwinter, colonies rely on stored honey and pollen which is contaminated with the ubiquitous neonicotinoid insecticide thiamethoxam, at mean concentrations from 6.4-28.9 ng/g.1 To determine whether dietary thiamethoxam affects overwinter survival of *Apis mellifera L.*, we chronically exposed winter bees to thiamethoxam in the field and in the laboratory and monitored survival. Field colonies chronically exposed to 100 ng/g thiamethoxam in fall sucrose syrup feeding experienced 65% overwinter mortality, which was significantly greater than the overwinter mortality of control colonies (10% overwinter loss, $P < 0.001$) and colonies exposed to 20 ng/g thiamethoxam (25% overwinter loss, $P = 0.011$). Under laboratory conditions, winter adult workers chronically exposed to 20 or 100 ng/g thiamethoxam in sucrose syrup had a significantly ($P < 0.001$) increased risk of death compared to control. Taken together, these field and laboratory results suggest that high environmental (20 ng/g) doses of thiamethoxam may decrease overwinter survival of honey bees and honey bee colonies. Further investigation of chronic exposure to mid-range environmental doses (5 and 10 ng/g) of thiamethoxam on overwinter survival is in progress.

[P.03.80] Effect of pesticides and honey bee pathogens on expression of immune related genes

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Intensive agriculture depends heavily on use of pesticides and because of their side effects they have become a major concern to a beekeeping industry. The ingestion of pesticides significantly alters the gut microflora and affect development, metabolic pathways, and immune defense mechanisms of honey bees. Furthermore, honey bees health can be additionally affected after exposure to different stressors, including pathogens (Varroa destructor and Nosema ceranae). In our studies, the combined effects of two stressors on honey bees in environmentally realistic conditions were investigated. We carried out experiments where honey bee larvae were treated with different pesticides (imidacloprid, thiamethoxam, prochloraz and coumaphos) and/or infected with pathogens (Varroa destructor or Nosema ceranae). We observed that different pesticides activate diverse expression patterns of tested genes, which depend on honey bee developmental stage and presence of different pathogens and parasites. Two pesticides, thiamethoxam and prochloraz, have different impact on gene expression of antimicrobial peptide Lysozyme-2. In honey bees exposed to thiamethoxam lysozyme-2 expression was downregulated, whereas in bees exposed to prochloraz expression of the same gene was upregulated through entire experiment on adult honey bees. Thiamethoxam or imidacloprid treatment also induced downregulation of the most immune related genes in late developing stages (white-eyed and/or brown-eyed pupae). In all studies honey bees were also tested for 10 most known pathogens. We observed that all honey bees exposed to varroa had significantly elevated Deformed Wing Virus (DWV) loads. It was also evident that most of the time response to different stressors varies depending on honey bee age. In our current and also our previous studies, we demonstrated that some pesticides can exert a suppressive effect on bee immune system. Simplified model to demonstrate interaction of pesticides and/or pathogens for bee colonies survival is not possible. We will discuss that specific mechanisms in honey bee colonies have to be noted according to the chemistry of pesticides and pathogen origin together with effects of the environment.

[P.03.81] Short-term exposure of thiamethoxam + lambda-cyhalothrin negatively affects the survival of Africanized *Apis mellifera* workers

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The honey bee *Apis mellifera* L. (Hymenoptera: Apidae) perform an important pollination service in the crops, visiting several plant species and ensuring the production. However, during foraging, the bees are exposed to pesticides sprayed in the crops. The excessive and, sometimes, incorrect use of these products, especially of synthetic pesticides, probably is one of the factors of colony decline, called Colony Collapse Disorder (CCD). In this sense, the objective was to evaluate the survival of *A. mellifera* when exposed to different concentrations of the commercial synthetic insecticide thiamethoxam (neonicotinoid) + lambda-cyhalothrin (pyrethroid). Treatments were insecticide thiamethoxam + lambda-cyhalothrin at concentrations of 100%, 80%, 60%, 40%, 20%, 10%, 5% of the recommended active ingredient for control the most important bug of soybean in Brazil, *Euschistus heros* F. (Hemiptera: Petatomidae). The control consisted of sterilized distilled water. Five replicates were performed with 20 workers each. Two bioassays were conducted, 1) contact of *A. mellifera* workers, for 2 hours, using Petri dishes (150 mm Ø) sprayed with solutions (290 μL/ dish) and 2) spraying the solutions (290 μL/ 10 honey bee) directly on recently emerged *A. mellifera* workers. After 2 hours of contact, the bees were placed in special cages and kept in a climatized room, being evaluated from one to 240 hours. However, at the end of the first two hours of contact with the product, it was observed that the bees of the treatments (bioassays 1 and 2) with the pesticide did not move in the cages, but they presented some movements. On average, 6 hours after contact with the products, all of workers death. Mouthparts externalization and subsequent darkening of the insects were observed as a toxic effect of the pesticide. Workers from the control had 100% survival after 200 hours. These results confirm the high toxicity of this product and alert growers that the user should be avoided at a pollination period not only in soybean but also in other crops where this pesticide is used. Studies are been performed evaluating the persistence and repellent effects of this product in honeybee workers in the laboratory and in the greenhouse.

[P.03.82] Use of the oxidative stress as a promising tool for biomonitoring the honeybee expositions to the pesticides

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The aims of the study were to compare the bioaccumulation of pesticides in beeswax and the impact on bee health through via the level of oxidation of proteins.
In French Guiana, three apiaries with three beehives of Apismelifera have been followed forone year. These apiaries were in three different situations of pesticides exposition: in the forest, as a control, close to a sustainable crop and the last one, close to a conventional crop. Sampling and honeybee preparation were done during dry and rain period following the AFNOR X43-909 normalization. The level of oxidative stress was monitored through the level of Carbonyl-SCORE (technology SDS PAGE patented).
As expected, the bee bread from control apiary (in forest) have less pesticides residues than in the two others exposed apiaries. Five

residues of pesticides were found in beeswax from the control apiary. The apiaries exposed in sustainable and the conventional crop area show respectively eight and seven residues of pesticides in their bee bread. Two pesticides recover were used as acaricide against Varroa destructor by the beekeeper (fluvalinate and coumaphos).
The level of protein oxidation was statistically more important (test: analysis of variance; p=0.0015) in conventional, than sustainable crop and control (forest), respectively 0.828, 0.617 et 0.465 carbonyl score unit. The level of oxidative stress was more important during rain period rather than dry period for all the apiaries.
This is the first study which describes the use of oxidative stress (protein oxidation) of potential tool for bee health survey.

[P.03.83] Reproductive fitness of honey bee queens exposed to thiamethoxam during development

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Productivity and survival of the honey bee colony depend on the reproductive fitness and health status of the honey bee queen. Poor queen quality has been reported as one of the main reasons of colony losses. Several studies have shown that neonicotinoid insecticides negatively affect reproductive potential of honey bee queens; however, the mechanisms and extent of this effect are not well understood. The aim of this study was to investigate the reproductive fitness of queens exposed to neonicotinoid thiamethoxam (THI) during late larval and pupal development.
On day 7 post oviposition, the grafted queen larvae were exposed to 4µl of distilled water containing 0 (negative control), 5ng (the highest estimated environmental exposure dose), or 50ng (positive control) of THI. The experiment was carried out once in June, 2018 and once in August, 2018. The following parameters were evaluated: larval and pupal survival, mating success, mated queen weight, sperm quality, and mandibular gland size.
Survival of queens until day 14 of development was 100% in the control group. Queen survival was reduced to 30.6% and 45.8% in the THI50 group in both studies (p=0.001 for both). Survival of queens in the THI5 group was only decreased in the August experiment to 68% (p=0.009) but was not affected in the June experiment.
No treatment effect was observed on total sperm count; however, sperm viability was decreased in queens exposed to THI50 by 18.4% ($p<0.001$) compared to control. Mandibular glands in both the THI5 and THI50 treatments were reduced by 12% (p=0.1) and 26.2% ($p<0.001$) respectively. Our study demonstrated that reproductive parameters of queens exposed to 50 ng of THI were negatively affected. The effects on reproductive fitness of queens that were exposed to the highest estimated environmental dose were variable between experiments conducted at different time points of the year.

[P.03.84] High susceptibility of North African honey bees to pesticides

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Apis mellifera intermissa is one of the numerous subspecies of the widely spread western honey bee *Apis mellifera*. It is native to North Africa and adapted to high temperatures, what makes it of a big interest regarding the current climate change.
Contrary to the European subspecies, the number of studies on the toxicity of insecticides on this subspecies is limited. Therefore, this study aims at evaluating the toxicity of thiamethoxam (neonicotinoid) and spinosad (biopesticide) to workers of the local honey bee *A. mellifera* intermissa.
The oral toxicity at 24h of thiamethoxam (Actara 25 WG, 25% of active ingredient (a.i.); 250 g (a.i.)/kg, water-dispersible granule, Syngenta Crop Protection) and spinosad (Tracer 480 SC, 480 g (a.i.)/l; suspension concentrate, Dow AgroSciences) (LD50 24h) were determined on newly emerged honeybees from untreated beehives in Annaba, Algeria where both insecticides are used for crop protection.
The LD50 by ingestion of thiamethoxam and spinosad at 24 h were 2.48 ng(a.i.)/bee and 192 ng (a.i.)/bee respectively (LC50: 0.31 ng(a.i.)/µl and 24 ng(a.i.)/µl respectively). It is lower than the LD50 values obtained for the European subspecies under similar methodological conditions for both insecticides. This implies that *A. mellifera* intermissa might be more susceptible to thiamethoxam and spinosad than the European honey bees upon which, recommended doses for crop protection are determined.

[P.03.85] Can honey bees detoxify Neonicotinoids

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Canadian honeybees have experienced high colony mortality in recent years with neonicotinoid usage on crops being a major contributing factor. In 2018, the Canadian Association of Professional Apiculturists reported that the national winter loss of bee colonies was 32.6%. Neonicotinoids are a class of agricultural pesticides that are chemically similar to nicotine. They are highly agonistic to insect nicotinic acetylcholine receptors (nAChRs), compared to the mammalian nAChRs. Honey bees tend to be more sensitive to N-nitro neonicotinoids (imidacloprid, clothianidin, thiamethoxam) compared to the N-cyano class (thiacloprid) due to the latter being more easily metabolized by the honeybee. Here we examine if honey bees that survive exposure to the N-nitro neonicotinoid clothianidin have different levels of expression of specific detoxification enzymes (e.g. CYP9Q1-3, a family of cytochrome p450 monooxygenases responsible for metabolizing neonicotinoids) relative to honey bees that die after exposure to clothianidin. We compared the expression of CYP9Q genes of bees of different genotypes after feeding them a field realistic dose (4.6 ppb), as well as an LD50 dose (29 ppb) of clothianidin.

[P.03.86] Neonicotinoids and other pesticides in honey produced in Jalisco, Mexico: analysis of environmental bee exposure and potential hazard to human health

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Neonicotinoid insecticides (NI) have been used worldwide since the 1990's. There is evidence of NI environmental contamination due to their systemic mode of action and persistence in different substrates. NI have been detected in food and human tissues. Additionally, the use of past generation pesticides, contributes to a toxic cocktail, threatening biodiversity, ecosystems functioning, and provision of services essential to human health, including food production that relies on pollinators, like the honey bee. Jalisco is Mexico's leading state for food and honey production, which has seen a 15% decline in recent years. It is suspected that NI have played a role in this decline. Lack of studies on the impact of NI and other pesticides on Mexico's environment and on the indirect effects on human health, impedes assessments based on scientific evidence. We propose that honey can be used as an indicator of environmental quality by determining the presence and levels of NI and other pesticides. Honey samples were collected from 30 different locations in Jalisco, Mexico. The samples were collected during the harvesting season. The samples were analyzed by LC-MS/MS multi-residue screen for NI and other 144 pesticides in the Agriculture and Food Laboratory from the University of Guelph in Canada. At least 19 pesticides were detected in 63% of the samples, 47% of them were imidacloprid and acetamiprid (NI) in combination with organophosphates, carbamates, fungicides and/or herbicides. This indicates that larvae and bees in hives have been exposed to pesticides. Honey's residue level of NI and 19 pesticides found in this study were compared with parameters of the EU LMR Codex Alimentarius to infer environmental and human exposure. This study improves our understanding about the cocktail of pesticides, NI in particular, that honey bee are exposed to and how this represent a potential hazard to human health.

[P.03.87] Microencapsulation of Clove Oil Using Spray Dry with Casein Micelle and Activity Test as Biopesticide Toward *Apis mellifera* bee

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Clove oil originates from leaves, flowers, and trunks of clove plant (Syzygium aromaticum) that is volatile and prone to both temperature and light. This research aims to decrease the rate of clove oil evaporation by making it into a microcapsule. This research starts with the microencapsulation of clove oil with a spray dryer using a casein micelle as a coating. Microcapsule resulted is evaluated in both physically and chemically as it is also tested for biopesticide activity towards *Apis mellifera* bee. This research has produced micro eugenol and micro casein products using spray drying with particle size 1,7162 - 2,0376 µm. The loading capacity of eugenol micelle casein increase with spray drying equal to 57,27% with an efficiency of encapsulation 87,99%. Encapsulation of eugenol with casein micelles using dry spray drying showed significant ability with the highest percentage change in mass of 9.81% at eugenol release rate. Biopesticides activity test towards *Apis mellifera* bee showed toxicity results in LD50 micro eugenol was lower when compared to LD50 eugenol. This study suggests that micro eugenol from clove oil has the potential candidate to develop as biopesticides toward *Apis mellifera*.

[P.03.88] Effects of thymol and carvacrol on carnolian honeybee after prolonged oral treatment: a comparison

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Monoterpenoid thymol, a component of essential oils, is widely used in beekeeping for varroa mite control, but it has negative effects on honeybee health. Another monoterpenoid carvacrol, found in high concentrations in oregano essential oils, has also strong acaricidal properties. Carvacrol could thus be promising for a regular use in beekeeping, however, its impact on honeybees has been poorly investigated. In this study we examined and compared the effects of 7 days oral treatment of carnolian honeybee workers (*Apis mellifera* carnica) with different concentrations of thymol and carvacrol on mortality and feeding. We also evaluated the activity of neuronal enzyme acetylcholinesterase involved in the control of the neurotransmission by hydrolyzing the neurotransmitter acetylcholine, and on detoxifying enzyme glutathione S-transferase in honeybee heads and thoraces. We show that both, thymol and carvacrol, caused significant mortality only at the highest concentrations tested, 1% and 5% respectively. As evidenced from the literature both substances could be effective

against varroa mites already at ten times lower concentrations, however, we found out that the 0.05% carvacrol or thymol exposure concentrations resulted in sublethal effects. They led to the increased activity of acetylcholinesterase and glutathione S-transferase in honeybee heads and thoraces. We conclude that prolonged treatment not only of thymol but also carvacrol could have adverse effects on honey bee nervous system resulting in a limited use for acaricidal purposes. We estimate that under chronic exposure conditions the toxicity of carvacrol is similar to thymol.

[P.03.89] Esterases Isoenzymes Expression in *Scaptotrigona bipunctata* Stingless Bees Contaminated with Fungicide

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Scaptotrigona bipunctata are stingless bees found in several regions of Brazil. Habitat fragmentation, loss of native vegetation and climatic changes are factors that contribute to decreasing populations of native bees. However, the excessive application of pesticides and the unsustainable use of agricultural ecosystems have been considered the main causes of diversity losses. Esterases isoenzymes act in the detoxification of xenobiotics in the organism of these insects. In S. bipunctata five esterases (EST-1 to EST-5) loci were described. Changes in their relative activity may indicate contamination with pesticides. Locker® is a fungicide that acts systemically or by contact and has triple action mode: inhibitors of tubulin (carbendazim), ergosterol biosynthesis (tebuconazole) and inhibitors of electron flow in mitochondrial respiration (kresoxim-methyl). This research aimed to analyze the esterases isoenzymes expression in S. bipunctata when contaminated by ingestion with three different concentrations of the Locker® fungicide. Adult foragers of S. bipunctata were collected from six colonies at the Experimental Farm Iguatemi (FEI) of the Universidade Estadual de Maringá, Brazil. Bioassays were performed with three replicates and one control group in a BOD chamber at 30 ± 2 ° C and 80 ± 10% relative humidity. Locker® fungicide contamination was achieved by ingestion for 24, 48 and 72 hours. Head/thorax of workers contaminated with Locker® and from the control group was prepared for PAGE electrophoresis and staining for esterases using A and B-naphthyl acetate. At 4.25 milligrams of active ingredient per millilitre (mg a.i./mL) partial inhibition of EST-4 (cholinesterase) was detected after 24 hours. Total inhibition of EST-4 was observed at 1.7 mg a.i./mL after 72 hours of contamination and at the concentration of 3.4 mg a.i./ mL after 48 and 72 hours. Other esterases showed no altered relative activities. In this way, we can conclude that Locker® is extremely toxic to S. bipunctata bees, evidenced by the higher mortality in 24 hours (25.35%) of intoxication.

[P.03.90] BeeSafety India: Creating a Mobile Phone App to Protect Pollinators from Pesticides in India and the Region

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When growers have access to information comparing pesticide toxicity to bees, they are empowered to choose products that protect their crop while posing the lowest risk to pollination services. Additionally, beekeepers are better able to communicate with growers about the timing and placement of bees relative to schedule and nature of pesticide applications. Oregon State University Extension Service has published such a resource, PNW591 How to Reduce Bee Poisoning from Pesticides, which was recently adapted into a mobile application. However, this app is targeted to pesticides, crops and beekeeping practices in the Pacific Northwest of the United States, which limits its usefulness in other regions of the world.

To rectify this deficit, Oregon State University and FoodPrint, an Indian NGO which promotes and advocates sustainable production practices in farming, are collaborating together to create an app similar to PNW591 for India and the region.

The app uses bee precautions from US and European products to characterize Indian pesticides as “highly toxic”, “toxic”, or “no precaution” for bees. Indian pesticide labels include few if any environmental precautions, and creating a BeeSafety India App may be the only comprehensive source of information Indian growers and beekeepers can access that will enable them to protect native and managed pollinators from pesticides.

Crop certification schemes provide sustainability standards for farmers and also open access to markets for their produce. Pollinator conservation is an essential element of sustainability, but not always part of certification programs. By partnering with crop certification programs to promote the app, farmers will be able to meet BeeSafety criteria. Food and beverage corporations who buy farmers’ produce could then claim that their products/inputs were produced in a bee friendly way as a part of overall sustainability. In this way, use of the app could provide more market access for farmers, protect bees from toxic pesticides, and benefit beekeepers’ livelihoods.

The BeeSafety India App provides a roadmap for similar country and region-specific apps that benefit farmers and protect pollinators around the world.

[P.03.91] Midgut Morphology of Stingless Bees *Scaptotrigona bipunctata* Contaminated with Acephate

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Scaptotrigona bipunctata is a stingless bee widely distributed in Brazil. Among the reasons that threaten bees are the loss of habitats,

high level of parasites and intensive use of pesticides. The main function of pesticides is to protect agricultural crops from diseases and pest. However, its use can cause negative effects in non-target organisms, such as bees. Organophosphate is a type of insecticide that causes severe damages to the nervous system, acting on the phosphorylation of the enzyme acetylcholinesterase, preventing its catalytic activity. This study used an organophosphate insecticide known as acephate, which inhibits acetylcholinesterase. When ingested by bees, the insecticide is metabolized in their midgut, therefore, morphological analysis is efficient to detect contamination by pesticides. In this research, we carried out an analysis of the midgut internal morphology of stingless bees S. bipunctata contaminated with organophosphate acephate. Adult foragers of S. bipunctata were collected from six colonies at the Experimental Farm Iguatemi (FEI) of the Universidade Estadual de Maringá, Brazil. The organophosphate insecticide used was Orthene® 750BR at the concentrations of 5, 7 and 9 micrograms of active ingredient per millilitre (μg a.i./mL). The bioassays were performed in three replicates, with a control group in a BOD chamber at 30 ± 2 ° C and 80 ± 10% relative humidity. Acephate contamination was achieved by ingestion for 24 and 48 hours. After this, the midgut of surviving bees was dissected for conventional histology and stained with hematoxylin and eosin. After 24 hours of intoxication, several alterations were observed, such as epithelial disorganization, increased microvilli, cellular degeneration, intercellular spaces, rupture of the musculature, detachment of the epithelium from the basal lamina and absence of the peritrophic membrane. After 48 hours, there was a reduction in the nest of regeneration, restructuring of the peritrophic membrane, loosening musculature and a slight detachment of the epithelium in relation to the basal lamina. In conclusion, the insecticide acephate is extremely toxic to S. bipunctata, evidenced by the higher mortality rate (15.5%) in the first 24 hours. After this period, the midgut morphology started a cellular recovery, contributing to the survival of these bees.

[P.03.92] Sulfoxaflor, a new enemy of bees?

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Over the last decade, large scale losses of managed honey bee colonies have been reported worldwide. The exposure of bees to different pesticides has been proposed as an important contributor to such losses. Sulfoxaflor is a new insecticide used to control aphids in crops. Although it is classified as highly toxic for bees, it has recently been approved in Uruguay for its use on soy, oats, ryegrass, apple and pear trees, wheat and barley. In this study, we aimed to evaluate the effect of sulfoxaflor on Uruguayan (Africanized) bees. Firstly we assessed the acute lethal dose 50 (LD50) of sulfoxaflor, according to EPPO´s guidelines. LD50 was estimated at 0.026 μg per bee, 6 times lower than the reported values (0.146 μg per bee). Then, we evaluated the effect of chronic exposition to sub-lethal doses (0.5, 1 and 2 μg/ml, intended to be 0.005, 0.01 and 0.02 ug/bee/day) according to OECD guidelines, for 24 days instead of 10. The lethal time 50 (LT50) was estimated in 3, 14 and 17 days for 0.5, 1 and 2 μg/ml, respectively, and was significantly shorter than in controls (22 days). Those results confirm that sulfoxaflor is highly toxic for bees, even at low concentrations. These findings confirm the importance of the evaluation of chronic exposure for long periods.

[P.03.93] The corn pollen, an important source of nutrients for honeybees and neonicotinoids residues

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The corn (Zea mays) is primarily cross-pollinated (95%) and needs vast amounts of pollen transported within a field to have an efficient pollination. Even the pollen morphology reflects an adaptation to wind pollination, its nutritional properties makes it a very important attraction for honeybees and other pollinating insects. Its male flower (the tassel) offer a large amount of pollen which shows that can be very attractive also in the period of sunflower honey flow. Some studies show that the crude protein in corn pollen can vary between 20-28% and in sunflower can vary between 15-18%. Other nutritional components in corn pollen can make it more competitive over sunflower pollen. This source of pollen is of great importance as the flowering period of corn is overlapping on the winter bees rearing so it contribute substantially to the quality of wintering, both by honeybee quality as well as by pollen storage which to be consumed in the early stage of brood rearing in the next season. The contamination of corn pollen with neonicotinoids may have a great negative impact on honeybees. Our studies show that all the samples of corn pollen collected in Romania contained different neonicotinoids in various proportions. The results will be analysed and discussed in the present study.

BEE HEALTH

10 SEPTEMBER 2019

POSTER SESSION 09 08:30-18:00
BEE HEALTH II POSTER AREA

[P.09.183] Whole-genome analysis of uncapping behaviour of individual honey bees towards Varroa destructor-parasitized brood

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Hygienic behaviour, i.e. the detection and removal of diseased brood, is seen as a central element of *Apis mellifera*'s social immunity with regard to brood parasites such as Varroa destructor. Hygienic behaviour is a heritable trait which can be used in selective breeding. In this study we aimed to elucidate and identify genetic variants related to this defence behaviour, with the aim of enabling marker assisted breeding. Worker bees from four honeybee subspecies (A. m. carnica, A.m. mellifera, A. m. caucasica and A. m. macedonica) and two crosses (MacedonicaXCarnica and MelliferaXCarnica) were individually screened for the expression of the behaviour in infra-red video observation hives with artificially Varroa-infested brood. Individual phenotyping allowed us to identify pairs of sister bees that either showed or did not show the trait of interest, which greatly reduces the background noise inherent to colony-level genomic comparisons. The different pools of “beginners” (those bees initiating uncapping of infested cells), “helpers” (continuers), “super-beginners” (involved in uncapping of more than one infested cell) and controls (observed at infested cells, but not involved in uncapping of Varroa parasitized brood cells) were subjected to whole-genome sequencing in order to identify SNPs, CNVs, and INDELs significantly associated with the behaviour. 3450 SNPs, 629 INDELs and 2061 CNVs were detected in regions significantly associated with the trait. Genotyping was performed in order to validate this association, which resulted in the confirmation of 42 genomic intervals, encompassing 167 genetic variants with high association with hygienic behaviour. In-depth functional annotation revealed several genes involved in associative learning, olfactory learning, as well as visual and neuroactive interaction. These results open a new perspective on the mechanism of hygienic behaviour and are presently being used to create an effective tool for the selection of varroa-resistant bees.

[P.09.184] Genetic parameters of two Swiss *Apis mellifera* populations and their implication for selection

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Genetic evaluation of selection candidate is a key step to determine the mating plan in breeding programs. In the honeybee, calculation of breeding values is performed for several phenotypes in different selection programs but few is known about the correponding heritabilities. Here, we applied a Best Linear Unbiased Prediction (BLUP) to an animal model to estimate breeding values and heritabilities corresponding to phenotypes measured by beekeepers in two selection programs in Switzerland. Heritabilities could be calculated for honey production, hygienic behavior, calmness and gentleness of the honeybees. Few variability of genetic origin was detected for criteria related to the infestation level of the parasitic mite Varroa destructor. In the Swiss context, with high regional colony densities, differences between colonies for these traits may be mainly due to horizontal between-colony mite transfers occurring due to drift or robbery. Implications for the selection programs are discussed.

[P.09.185] IHBBN, reducing colony losses by breeding locally adapted honey bees

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Nowadays the Western honey bee (*Apis mellifera*) is present worldwide, wherever beekeeping is possible, though its native habitat spreads out across Europe, Africa and parts of Western Asia. About 26 different subspecies evolved through time, each adapted by natural selection to cope with the local environmental conditions. During the past 2 centuries, Western honey bees were transported from Europe to the Americas and Australia. Modern beekeeping increased inbreeding of some honey bee populations within the native

range as well, with a few favoured subspecies being imported into natural habitats of other subspecies. Some subspecies, such as *Apis mellifera* adami, are almost extinct while of some others, such as A. m. siciliana and A. m. ruttneri, there is just a handful of populations remaining, needing intensive conservation measures. Regardless loss of biodiversity, there are indications that genetic pollution could lead to maladaptation to local environmental conditions, making it harder for honey bee colonies of non-local origin to survive. The E.U. funded project SMARTBEES was also aiming to develop breeding activities all over Europe and especially breeding for traits important to beekeepers and mite tolerance, and breeding in countries with endangered and neglected native honey bee populations. These breeding activities would hopefully lead to conservation through utilization. The SMARTBEES work package responsible for this breeding extension became a great success and when the project finished, the participants agreed there was continuity needed to keep the breeding efforts going. Therefore, the International Honey Bee Breeding Network, IHBBN, was established. IHBBN is an association, set up by associations and groups involved in SMARTBEES, aiming to outreach to breeding associations worldwide, which has set itself the goal to encourage and support breeding locally adapted honey bees, in their native range where it could help conservation of endangered subspecies, but also outside the native range where however the benefits of local breeding can also be present. As well as for the Western honey bee, this assistance will also be offered for the Eastern honey bee (*Apis cerana*).

[P.09.186] Software to facilitate estimation of genetic parameters and breeding values for honey bees

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Wageningen University & Research, Animal Breeding and Genomics, Wageningen, THE NETHERLANDS

Effective breeding for desired traits requires knowledge of genetic parameters, such as heritabilities, and breeding values. Theory and methods required for bee populations have been developed by Bienefeld et al. (Apidologie, 2007, 38:77-85), Brascamp and Bijma (Genetics Selection Evolution 2014, 46:53-67) and Bernstein et al. (J Anim Breed Genet. 2018, 135:323-332), and software is available from the field of animal and plant breeding (e.g. Gogel et al, ASReml 4.1, 2015, VSN International Ltd, Hemel Hempstead, HP1 1ES, UK). However, practical implementation in honey bees is challenging because: 1. Observations are not on single individuals but on colonies; 2. the sire of workers (the mate of the queen in the colony) usually is not a single individual.
We developed software that supports the practical implementation of the estimation of genetic parameters and breeding values for honey bees. The software uses inputs that are routinely available in most bee breeding programs, and provides output files that allow straightforward application of standard software packages for genetic analysis (such as ASReml). The software consists of two programs in R. The first program builds a pedigree, using the identification of the queen in a colony, her dam, and the mate of the queen plus birth years. Mates can be one queen, a group of sister queens as well as queens in open mating. The numbers of queens are input along with the mean number of fertilizing drones for a particular mate. A pedigree always has sires without known pedigree. For these sires the software allows different assumptions for the mutual relationships of the queens involved (Brascamp & Bijma, 2019, submitted).
The programs have been used in several (collaborative) projects. The programs are available on request, and are still regularly updated and extended.

[P.09.187] Honeybee Regeneration Project

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The Honeybee Regeneration Project is a project we are working on to look at ways to set-up breeding programs, create more resilient and localized stock, and then share breeding knowledge with other beekeepers. The project was made possible by a SARE grant.
Goals:
1) Design a breeding program to create more resilient honey bee stock.
2) Apply the breeding program to our own stock in our local environment
3) Share both the honey bee stock we've created, as well as the guidelines for creating a similar breeding program, with other beekeepers.
Beekeepers are living in a perilous time. With high pressures from mites and pathegons it has become increasing difficult to keep hives healthy and striving. Many beekeepers believe that the key to many of the problems bees face is hidden within their own DNA. For years I had been rearing queens and only slighting embarking upon the actual practice of conscientious breeding. As I looked deeper into the realm of bee breeding I realized there were many resources unavailable to prospective breeders. At this point I decided to create a breeding program and protocol, monitor the accomplishments and setbacks, and then give back to the beekeeping community with the lessons I have learned.
In spring 2018 we began by selecting 5 breeder queens. We then grafted 10 daughters from each breeder. Over the course of the last year we have monitored for mites and disease, productivity, temperament, brood pattern and general health and well being. From the top performing 5% of those queens we have selected our 2019 breeders. Daughters of those breeders we have sent out to other queen producers so that they may analyze them in their own operations.
At the heart of this project is outreach. We have been sharing what we have learned with other beekeepers via presentations, field days, classes, slideshows, blogging, video and social media posts. We hope that other small-scale producers and beekeeping cooperatives may use the knowledge we have attained to set up their own breeding programs and improve their own localized stock.
You can find more information here:

[P.09.188] Models of Community Breeding Projects

B. Morse
Bee Audacious, San Rafael, USA

A local approach to addressing honey bee health: In Marin County, California, sourcing bees locally increased from a reported 42% to 85% between 2009 and 2015. At the same time, reported losses decreased from a high of 53% to a low of 34%. This was accomplished through a variety of efforts including: 1) education (including both annual census survey showing beekeeping practices and associated loss rates to encourage changes in management and increasing educational level of local beekeepers on queen rearing and making increases); 2) prioritizing stock selection and breeding, and 3) community outreach. Regardless of whether beekeepers had 1 hive or 50+, all were encouraged to be part of the working solution to counteract high loss rates in the county. This collaboration has enabled us to continue to work collectively to address additional challenges that have emerged in recent years including the introduction of small hive beetle in the county and the changing challenges associated with increasing hive densities.

[P.09.189] Biochemical and molecular characterization of a striking coumaphos resistance mechanism in Varroa destructor

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Varroa destructor and the viruses it transmits is one of the most important problems in modern beekeeping worldwide. The use of chemical acaricides has been the main practice to control these mites since their introduction in Europe and the US. Although more alternative control strategies become available, chemical control of Varroa continues until today in many regions of the world.
One of the main acaricides used for Varroa control is the organophosphate coumaphos, which has excellent selective toxicity. The frequent use of this acaricide has in some regions placed a strong selection pressure with the development of resistance as a consequence.
We report here on a Varroa destructor population that exhibits extreme high levels of coumaphos resistance. Sequencing the acetylcholinesterases gene, the target-site of coumaphos, did not reveal any resistance mutation, nor did biochemical tests revealed different binding kinetics, thus excluding target site resistance. Coumaphos is metabolized to coumaphos-oxon the active principle, but chlorferon is considered the main non-toxic metabolite. Although we could clearly identify chlorferon as the principle metabolite after incubation with radio-labeled coumaphos, we could not detect different rates of metabolite formation between susceptible and resistant Varroa destructor strains. Surprisingly, the amount of coumaphos-oxon was barely detected in resistant mites. All data together points towards a novel resistance mechanism, a hypothesis supported by deep RNA sequencing and functional assays.

[P.09.190] Should Varroa Sensitive Hygiene behaviour be included in a sustainable beekeeping program?

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The ectoparasitic mite Varroa destructor Anderson & Trueman (Acari: Varroidae) causes major damages and economic losses to the beekeeping industry. Several acaricides have been successfully used over the last decades to control Varroa infestations, but their repeated use has led to the development of pesticide-resistant mites. It is thus imperative to find new solutions to control this parasite. The selection of honey bees that express the Varroa Sensitive Hygiene behaviour (VSH) could be a potential and sustainable solution. Honey bees who express this behaviour are able to keep the populations of Varroa living in their hive under control by removing mites contained in brood cells. Nevertheless, selection of a specific phenotype carries a certain risk since it may have a negative impact on other desirable performance traits. Therefore, the purpose of our study is to determine whether a high VSH behaviour expression allows colonies to effectively control Varroa mites populations without affecting other important selected performance criteria in apiculture. To do that, sixty-one honey bee colonies were selected from the Centre de recherche en sciences animales de Deschambault breeding program. These colonies were evaluated and a regression analysis was carried out between VSH behaviour and various performance traits (honey production, spring development, defensive behaviour, hygienic behaviour, winter survival and winter feed consumption).

[P.09.191] Honey bee health is mediated by propolis production and consumption

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Bees produce propolis from resins that they collect from plants. Many of these resins have chemical properties that protect these plants against diseases. Bees recognize and take advantage of the antimicrobial activity of these plant resins to protect the integrity of their nest cavity and to help combat parasites and pathogens. The medicinal properties of propolis have been widely demonstrated and applied in human and animal health in some countries; however, its importance for bee health is not clearly established or taken advantage of. In fact, historically beekeepers have selected bees to produce less propolis because it blemishes comb honey, sticks to gloves and hands, and makes it difficult to manipulate frames and other hive parts. Current concerns about bee losses are a motive for reexamining how bees naturally deal with threats to their health. Africanized bees, which are quite resistant to bee parasites and pests, are generally prolific propolis producers. In Brazil, honey bee colonies are not treated for mites, hive beetles or any disease. Since propolis is a major commercial product in Brazilian apiculture, beekeepers in some regions of the country even select for increased production of this hive product. We have found that bees in colonies that produce more propolis have increased brood viability and adult worker lifespan. They also have more efficient hygienic behavior and larger honey and pollen stores. In an attempt to better understand the impact of propolis on bee health, we added 0.1% propolis to a pollen substitute diet and found that the expression of four key immune system genes was greatly increased in nurse bees injected with bacteria. We suggest that instead of constantly adding chemicals to colonies, more attention should be paid to how bees naturally deal with pests and diseases, including their production and use of propolis.

[P.09.192] Transcriptome analysis of selective bred varroa resistant honey bees: unraveling the molecular basis of varroa resistance

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Varroa destructor is causing considerable damage in bee populations in Europe and USA. Periodical Varroa treatments have become necessary in the survival of Western bee populations. These varroa control strategies increase the cost of bee-keeping, lack efficiency and often have a negative impact on the overall health of the bees and the risk of chemical residues in bee products. Selective breeding of honey bees for naturally occurring resistance behaviors can lead to efficient Varroa resistance (VR) and circumvent the need for anti-Varroa treatments. VR has been described as the capacity to detect, uncap and remove infested (or damaged) worker- and drone brood (hygienic behavior) in addition to the active cleaning and damaging of foraging mites from adult bees (grooming).

We have performed selective breeding (using artificial insemination) of a population of *Apis mellifera* Buckfast for the properties of (brood) hygiene and mite damaging auto- and allogrooming, which has allowed us to abolish anti-Varroa treatments for more than 3 years and maintain stable and healthy populations of VR Buckfast bees. These VR bees display very high hygienic behavior, with a capacity to detect and remove freeze killed brood in 100 cells in 9hrs. The varroa burden in these colonies is very low and following induced infection with Varroa the percentage of reproducing Varroa mites in VR-broodcomb was significantly lower as compared to broodcomb from non-Varroa-resistant (NVR) colonies. In addition to this phenotype we observe a high level of recapping occurring in these hives, suggesting the VR bees use recapping as one of the strategies to disrupt Varroa mite reproduction. Fallen mites from VR hives also show significantly more inflicted damage and VR bees display efficient auto- and allogrooming in response to external stimuli, indicating that hygiene towards brood and hygiene towards the bees themselves are two important aspects of varroa resistance. We performed transcriptome analysis of VR bees compared to NVR bees and have identified candidate genes that are associated with behavioral and olfactory traits that underly Varroa resistance. Following validation of these genes we provide a basis that allows to evaluate the level of Varroa resistance present in a bee population.

[P.09.193] Marker-Assisted Selective Breeding for Resistance Against American foulbrood (Paenibacillus larvae)

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American foulbrood (AFB) is a devastating brood disease of honey bees (*Apis mellifera L.*). Colonies with visible symptoms typically die if not treated, resulting in considerable economic loss to beekeepers. Relying on antibiotic treatment is problematic, due to the development of resistance and the potential to contaminate honey. A more sustainable approach is to select and breed bees with increased ability to limit or eliminate brood diseases. Bees bred for hygienic behaviour (HB) have the ability to detect and remove infected or dead brood from the colony, thus eliminating the spread of disease. We have previously reported the identification of protein markers that correlate with HB and their use in selective breeding. In this study, we demonstrated the utility of this proteomic tool to enhance disease-resistance over three generations. In the first year, we evaluated 635 colonies from four provinces in western Canada, for hygienic behaviour using the freeze-killed brood assay. The F0 population was established from hygienic colonies and a benchmark population was randomly selected. In the two subsequent years, we evaluated, selected, and propagated three successive generations based on either hygienic behaviour (Field-Assisted Selection, FAS), or protein marker selection (Marker-Assisted Selection, MAS). The performance of the two selectively bred lines, FAS and MAS, in F1 and F3, was compared with the unselected benchmark and a commercially imported stock. In each generation, colonies from all four stocks were challenged with *Paenibacillus larvae*, by introducing a 15 x 15 cm section of comb containing 30-50% cells with visible AFB symptoms.

Our results showed that by the F3, MAS was as efficient as FAS for selective breeding for HB as tested using a freeze-kill brood assay. Selectively-bred FAS and MAS stocks had fewer clinical symptoms of AFB as well as fewer colonies infected by the end of the season. Colony survival was also higher in selected colonies, compared with the benchmark and imported stocks. No difference was observed in honey production among stocks, indicating that selective breeding for HB using MAS improves resistance to disease without compromising colony productivity.

[P.09.194] Genetic basis of behavioral resistance to varroa mites in Carniolan honey bee in Slovenia for selection in the future

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Carniolan honey bee (*Apis mellifera* carnica) is autochthonous and protected subspecies in Republic of Slovenia. It is highly appreciated among beekeepers because of its favourable qualities such as extreme calmness, adaptation to low temperatures and good honey production. Infestations with varroa mite (Varroa destructor) are major problem in colony health and one of the main reasons for colony loss. It has been recognised that there are long-term benefits from selection of colonies that exhibit varroa sensitive hygiene. In Slovenian breeding program hygienic behavior as one of the components of social immunity has been monitored using the pin-test but the results are currently not included in selection of genetic material for breeding purposes. With exception of a few studies on population structure of Carniolan honey bee in Slovenia, genetic characterisation has not been employed. In 2017 we launched a project that aims to investigate genetic variation in Carniolan honey bee colonies from Slovenia and identify genetic markers that are related to hygienic behavior. In the first phase we have developed efficient noninvasive extraction of DNA from exuviae of queens' pupae which serves to determine desired genotype of selected queens before they are inseminated. This method enables establishment of faster selection of varroa resistant lines and assistance in breeding for desirable traits. Here we present our initial results and steps to be taken in next two years. We provide first insight into the selection of molecular markers and the variation of selected markers in monitored Carniolan honey bee populations in Slovenia. Our project sets basis for assistance in selection of other interesting traits in honey bees by noninvasive genotyping.

[P.09.195] Strengthening the commercial apiculture industry in Newfoundland, a mite-free honey bee population

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Newfoundland, one of two confirmed locations in the world free of Varroa destructor, could supply honey bee (*Apis mellifera*) nucs to mainland Canada and America. However, Newfoundland has some unique issues that need to be addressed before the commercial industry can compete in supplying nucs. A big issue is that drones become sexually mature much later in the season relative to mainland NA, thus, delaying the production of fertilized queens. My research focuses on two important areas: optimizing drone semen cryopreservation in order to maintain sperm viability into the next season and assessing the characteristics of the honey bee lineages on the island. Success with cryopreservation will enable the instrumental insemination of queens earlier than what naturally occurs. The current record is shy of a year with approximately ¾ viability obtained, but to be useful to the industry in Newfoundland it will need to be extended and will benefit greatly from increased viability. Using these methods as a control, I will vary cryoprotectant amounts and freeze time to determine the best approach to sperm preservation. I will be presenting the initial viability measurements for this year long experiment. Additionally, by evaluating the current subspecies within a local apiary on the island I will put forth a recommendation on what lineage makes the most difference on increasing honey bee populations in Newfoundland. The island is home to the Italian honey bee [*Apis mellifera ligustica*], Carniolan honey bee [*Apis mellifera* carnica], and the uncontrolled mating of these two lineages [Newfoundland honey bee]. These queens will be instrumentally inseminated with drones of their own subspecies, drones from different subspecies, and a combination of all drone semen which will produce 12 different queen types. The colonies they establish will have the following hive characteristics assessed: gentleness, queen brood laying pattern, amount of honey produced, pollen sources, temperatures workers can forage in, and brood diseases. I will be presenting my findings on hive characteristics of the various queens and progeny from the selective breeding.

[P.09.196] Sacbrood Virus Resistance Honey bee (*Apis cerana F.*) in Korea

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South Korea has over 0.38 million of managed honey bee (*Apis cerana*) colonies before 2009 years ago, which produce the highest quantity of honey in the Korea; however, almost colony (90%) were collapsed by Korean Sacbrood Virus (KSBV) in South Korea. Korean Sacbrood Virus (KSBV) is the pathogen of *A. cerana* Sacbrood disease, which poses a serious threat to honeybee *A. cerana*, and tends to cause bee colony and even the whole apiary collapse. Colony collapse of *A. cerana* was first reported on the Pyeong-Chang of the South Korea in 2009. Several scientists and governments has been tried research for cure the sacbrood disease in *A. cerana* colony by medicines and management techniques. Unfortunately, The sacbrood disease dosen`t improve. So, we were developed a better breed of *A. cerana* for resistance of sacbrood virus by selection and then artificial insemination. *A. cerana* breeding technique was first successful applied with *A. cerana* in Korean. Queens was grafted from sacbrood resistance line and then it was growing in sacbrood disease colony that was survived 100%. Altogether selected 18 queens were artificially inseminated and 2,000 drones of *A. cerana* in Korea was used to evaluate amount of semen collection. We are select two scabrood resistance *A. cerana* line (R and H). R line be used for rearing the Queen. Drone was reared in H line colony. The RH hybrid were not infected sacbrood virus even spread sacbrood virus (2×106). RH colonies have very excellent hygienic behavior, brood, and sacbrood disease resistance activity.

[P.09.197] Effect of the contribution of sugar syrup on larval survival in bee colonies affected by River Disease

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The "River Disease" is an intoxication from a toxic honeydew that produces massive death of bee larvae. The objective of this study was evaluate the contribution of sugar syrup in 2 forms of availability as diluent of the honeydew entering to the colony. There used 30 bee colonies located in a place where naturally severe cases occurred in previous years. The colonies were divide into 3 groups, Control (C), Quick Availability (Q), and Slow Availability (S). In Q group is added an internal feeder to available all syrup within first 12 hours and the S Group was provide with a Boardman feeder where the syrup is available within 48 to 60 hours. Colonies were visit 5 times every 9 days. In the first 4 sessions was provide with 2 litres of sugar syrup 1/1 to the groups Q and S, to all colonies in each session were photographed one frame with eggs, which was again photographed in the next session, except in the 5th where only frames with eggs from the previous session were photographed. Image analysis was perform to estimate larval survival (LS) through ANOVA analysing. The LS in C was lower than 3% in all sessions, while Q and S always was find significant differences with C. When Q and S are compared within each session, no significant difference is found between groups, but when we compare them over the time; in the first 2 sessions LS is above 50%, and in the other 2 sessions it is significantly lower (between 24% to 38%). The offer of syrup in Q or S prevent most of the deaths that occur in C. The LS decrease in time, possibly because the effect of dilution occurs while the syrup is available and the other days the toxic honeydew continues entering and accumulating in the colony, so that more and more larvae die in time. We consider that provide sugar syrup is not a viable productive management, because it does not prevent the massive larval mortality in the time, in addition, sugar residues could be evidence in the honey.

[P.09.198] Integrated Varroa management: Compared expression of hygienic behaviors in two selected lines developing in Ontario and Quebec

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The parasitic mite Varroa destructor represents to this day the biggest threat in apiculture. A promising solution would be the genetic selection of hygienic behaviors, including Varroa Sensitive Hygiene (VSH) behavior. Indeed, the detection and removal of pupae infested with varroa females by bees that have the VSH trait makes it possible to limit population growth during the season, and therefore to increase resistance to the parasite. On the other hand, few studies have observed the effect of the external environment on the expression of this behavior within the colony. In addition, although the VSH behavior detection method developed by Büchler et al. (2017) is the most reliable so far, it is very heavy logistically and not very fast. That's why my study is comparing the performance of selected colonies for VSH and hygienic behavior in different environments, in Quebec and Ontario. On the other hand, I am verifying whether it is possible to use a more simple phenotypic test used for hygienic behavior by modifying it to make it specific for the detection of VSH behavior.

BEE HEALTH

11 SEPTEMBER 2019

POSTER SESSION 15 08:30-18:00

BEE HEALTH III POSTER AREA

[P.15.310] Impact of Varroa destructor and deformed wing virus on emergence, cellular immunity, wing integrity and survivorship of Africanized honey bees in Mexico

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The ectoparasitic mite Varroa destructor is the main health problem of honey bees (*Apis mellifera*) worldwide. Africanized honey bees in Brazil have demonstrated tolerance to the mite, but there is controversy about the degree of mite tolerance of Africanized bees in other countries. This study was conducted to quantify the effect of V. destructor parasitism on emergence, hemocyte concentration, wing integrity and longevity of Africanized honey bees in Mexico. Africanized bee brood were artificially infested with V. destructor mites and left to develop in an incubator until emergence as adults and compared to non-infested controls. Deformed wing virus (DWV) presence was determined in the mites used to infest the bees. After emergence, the bees were kept in an incubator to determine survivorship. The percentage of worker bees that emerged from parasitized cells (69%) was significantly lower than that of bees emerged from non-infested cells (96%). Newly-emerged parasitized bees had a significantly lower concentration of hemocytes in the hemolymph than non-parasitized bees. Additionally, the proportion of bees with deformed wings that emerged from V. destructor-parasitized cells was significantly higher (54%) than that of the control group (0%). The mean length of life of bees that emerged from infested and non-infested cells was 8.5± 0.3 and 14.4± 0.4 days, respectively, and the difference was significant. It is concluded that V. destructor parasitism and DWV infections kill, cause deformities and inhibit cellular immunity in developing Africanized honey bees and significantly reduce the lifespan of adult bees in Mexico. These results suggest that the tolerance of Africanized bees to V. destructor is related to adult bee mechanisms.

[P.15.311] Canadian National Honey Bee Health Survey 2014-2017

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In Canada, over winter losses were reported to be an average of 27.3% between 2007 and 2013, a level considered unsustainable and well above the range of 10% to 15% previously considered as acceptable. The Canadian beekeeping industry was concerned and showed increasing support for a honey bee health surveillance program. Between 2014 and 2017, the first Canadian National Honey Bee Health Survey collected 944 apiary level sample sets from 338 beekeepers across Canada. Samples were taken from mid-July to mid-August and analyzed for endemic and exotic health treats. In 2017, neonicotinoids residue analysis was performed in bee bread. Results indicate Nosema ceranae as the dominant species in all provinces, with single infections of Nosema apis becoming rare. Infestation rates of Varroa destructor varied across years with an average load of 1.5 mites per 100 bees. The prevalence of Nosema was significantly greater in migratory operations (69.0% vs. 56.7%), while Varroa infestation levels were higher (1.65 ± 0.14 vs.1.25 ± 0.15) in stationary colonies. *Paenibacillus larvae* was detected in 11.8% of the samples, and 26.8% of the isolated strains showed resistance to antibiotic tetracycline, but no cases of tylosin resistance were detected. The most prevalent viruses detected were Black Queen Cell virus, Deformed Wing virus and Sacbrood virus. Tropilaelaps spp. and Slow Bee Paralysis virus were not identified in any samples, providing empirical evidence to support Canada's zoosanitary status for the trade of bees with other countries. Although 88% of samples in 2017 tested positive for 1 or more neonicotinoids, on average, the levels detected were well below 1 ppb, which are not normally associated with lethal effects on honey bees. Results from this survey provide essential information on the baseline of honey bee pests, diseases and pesticides in Canada. A continuation of the program in the future can play an important role to maintain healthy, sustainable and profitable beekeeping in Canada.

[P.15.312] The dwarf honeybee as an introduced species into Taiwan, the eradication and study on the parasites on *Apis florea*

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The dwarf honeybee (*Apis florea*) is common seen honeybee species in Southeastern Asia. Since October 2017, it was found that the wild population introduced into Taiwan. In order to prevent threatens of non-native species to the ecosystems of Taiwan, the surveillance of occurrence for dwarf honeybees was done by our team supported by Taiwan government. Until now, at least nine colonies have been eradicated. The nest location, nesting habitat, colony size and composition were measured. The phylogeography analyzing suggests that the introduced population is closest to Thailand. The examinations of eradicated individuals, however, the ectoparasite mites (Euvarroa

sinhai) and pathogens of black queen cell virus, deformed wing virus and kakugo virus (KV) were identified. Here we provide new evidence that KV not only infect western honeybees but also dwarf honeybees. Overall, the introduced dwarf honeybee may be caused from the frequent international goods transportation. We are not sure the actual effect of Taiwan ecosystem or apiculture. The risk assessment of introduced dwarf honeybee must be further established.

[P.15.313] Underpinning the molecular mechanism of Ascosphaera apis pathogenicity to honeybee larvae and biological control to chalkbrood disease

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Ascosphaera apis is a chalkbrood causing pathogen in honeybees. Chemicals have been used against to this disease. For better understanding the pathogenicity of the A. apis and finding its biological control options we accomplished the following experiments: (1) optimized the protocol of protoplast preparation involved in A.apis. In this work, an efficient genetic manipulation of this fungus has been well developed and reproducible protocol to make this fungus amenable to genetic studies and transformation. (2) Based on the above efficient genetic manipulation of the fungus, Restricted Enzyme-Mediated Integration (REMI) was used to construct stable A. apis transformants to investigate the infectivity of transformed pathogens to honeybee larvae. 12 mutants were selected for further study to identify pathogenicity associated genes. In vitro bioassay confirmed that 10 of the selected mutants had notable differences in pathogenicity among themselves and with the wild-type strain. (3) pathogenicity tests have been conducted by artificial methods using spore inoculation and in vitro larval rearing techniques. (4) De Novo Sequencing and Transcriptome Analysis of A. apis in Response to Pathogenicity to Honeybee Larvae. We sequenced, assembled and annotated the transcriptomes of three mutants and wild-type of A. apis pathogens for comparative transcriptome analysis to better understand the gene expression regulation during sporulation. KEGG pathway analysis revealed that functional groups of differentially expressed genes associated with Proteasome pathway, Fatty acid metabolism, Glycine, serine and threonine metabolism, Aminoacyl-tRNA biosynthesis, Fructose and mannose metabolism, SNARE interactions in vesicular transport which play a vital role in virulence involved in this fungus. Six genes (OmtA, Ski3, TOXD, Hts1, Nor-1 and melanin) were highly expressed in SPE1 samples compared to its corresponding non-pathogenic SPE4 which had lost pathogenicity on honeybee larvae and had relatively the lowest expression levels of selected candidate genes compared to the wild-type. (5) Trichoderma transformants constructed by REMI could be used as potential biological agents against the growth of chalkbrood diseases in bee hives.

[P.15.314] When being lazy pays off: Different stressor interactions between drone and worker honeybees

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Interactions amongst stressors are suspected to be responsible for increased honeybee colony mortality. Although it seems likely that, e.g. poor nutrition would exacerbate the effects of a pathogen, few studies have actually measured the interaction effects and even fewer have addressed drones despite their key role for reproduction. Here, we tested the susceptibility of both honeybee workers and drones to the single and combined impacts of malnutrition and the common microsporidian pathogen Nosema ceranae. Drones were not significantly more susceptible to N. ceranae than workers as indexed by spore load and rate, body mass, and survival. However, workers were significantly more affected by malnutrition, having significantly reduced survival in all malnourished treatments compared to drones. Additionally, the data demonstrate distinct stressor interactions between the sexes. Whereas workers broadly showed synergistic interactions and increased stress, drones showed antagonistic interactions and decreased stress. It appears as if “cryptic” stressors not included in the study design may have contributed to these findings. Indeed, differential stress may have been imposed by the experimental design, e.g. lack of brood in the cages. Alternatively, but not mutually exclusive, life history traits could have contributed, e.g. young nurse workers are feeding drones and are therefore per se more physiologically active than the male sexuals. Future studies on stressors should not only take into account possible differences between species, but also between sexes and castes within single species as well as the actual nature of stressor interactions.

[P.15.315] Modeling the spread of Varroa resistance in feral colonies

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As Varroa destructor becomes endemic in an area the feral *Apis mellifera* population quickly dies and the free pollination service provided by them to horticulture is lost. Although the feral population of honeybees usually returns to the same numbers after about 10 years as Varroa-resistant bees are selected, an immediate shortfall in colonies for pollination is expected. We developed a model to determine if it is feasible to seed the managed population with Varroa-resistant, Varroa Sensitive Hygienic (VSH) bees as a way to accelerate the growth in resistance in the feral honeybee population.
Current models of the spread of Varroa resistance in honeybee populations are limited by the complexity and randomness of the mating process. In this stochastic model we assumed that each virgin queen mates randomly with 16 to 18 drones from other colonies. It is assumed

that some of these drones carried the VSH trait while others did not. Also, the virgin queen may have the VSH trait. A population of honeybees was modeled over 15 years and differing assumptions made about the survival of colonies depending on the proportion of workers that possessed the VSH trait. Both feral and managed populations were included in the model although these were kept separate apart from random mating of feral virgin queens with drones. It was also assumed that a proportion of managed colony swarms join the feral population. Depending on the proportion of workers in a colony that have the VSH trait and the assumptions made about the proportion of workers resistant versus the probability of colony death, it was possible to model feral population collapse and recovery. Our results are consistent with that which has been experienced in some survivor populations in Europe and North America.

[P.15.316] Why are we seeing such high rates of European Foulbrood disease after blueberry pollination, and what can be done?

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Beekeepers often report high rates of European Foulbrood after their colonies are on blueberries for pollination. This phenomenon has been reported all over the United States and Canada, and many anecdotal reports indicate that it has been worsening in the last few years. EFB can significantly weaken colonies, and reduce the potential honey crop. Because of the timing in early summer, beekeepers have to choose between using antibiotics to treat their hives, or to let the colony go, potentially missing in honey production.
Michigan is one of the top producing states of high bush blueberries, and beekeepers often report issues with European Foulbrood in their hives. In 2018, we began a program investigating the disease prevalence as well as risk factors that led to infection. We worked with commercial beekeepers to sample colonies at 14 blueberry fields. We examined pesticide exposure and nutrition as possible risk factors that could lead to disease. We estimated pesticide risk through landscape analysis, spray records, and testing incoming pollen. We identified nutritional risk factors by examining the protein of the incoming pollen as well as through an experiment with supplemental feeding. In 2018, we found high rates of EFB in colonies that were on blueberry pollination. The honey bee colonies had access to good amounts of wild pollen at that time, but we saw lower rates in colonies that were fed supplemental pollen substitute at that time.
We also followed the colonies through recovery, and did not find significantly different rates of recovery for colonies that had antibiotics, protein supplement, both antibiotics and protein supplement and control (nothing). Both of these studies will be continued in 2019 to provide two years of data on fungicide exposure and to replicated the cohort recovery trial.

[P.15.317] Investigating Incidences of European Foulbrood and Other Pathogens in Highbush Blueberry-Pollinating Honey Bees

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Blueberries are Canada's top fruit crop export, with cultivated blueberries occupying 75,000 hectares of land. In 2016, Canada produced more than 200,000 metric tonnes of blueberries, valued at $400 million. British Columbia produces the majority of Canadian cultivated highbush blueberries (Vaccinium corymbosum), which are mostly pollinated by honey bees originating from British Columbia and Alberta. Recent reports indicate a decline in honey bee colony health, particularly an increase in European foulbrood (EFB) frequency, after colonies return from pollinating blueberries. Our goal was to investigate the honey bee colony health status following blueberry pollination, with an emphasis on EFB infections.
For this study we examined five blueberry-pollinating apiaries and one non-blueberry apiary across three time points (start, end, and post-blueberry pollination). We observed a greater increase in EFB-like symptoms in the blueberry-pollination apiaries compared with the non-blueberry-pollinating apiary. Field observations showed all blueberry-pollinating apiaries experienced an increase in EFB-like symptoms following blueberry pollination. Laboratory analysis incorporated the novel technique of droplet digital PCR (ddPCR) to obtain absolute quantitation of EFB infection in honey bees across pollination time points. Molecular techniques confirmed field diagnosis of EFB-like symptoms in 77% of the samples. Colonies in blueberry-pollinating apiaries experienced a modest increase in Varroa infestations relative to the non-blueberry pollinating apiary, however Varroa levels did not increase in all blueberry-pollinating apiaries over time. We are also investigating Nosema spore counts and species to further evaluate honey bee health.
The observed increased incidence in pathogens following blueberry pollination indicates that beekeepers' observations merit further investigation to determine the exact variable(s) that are affecting bee health. These studies will guide management strategies to ensure the sustainability of healthy honey bee colonies that are able to continue pollinating blueberries and other agricultural crops.

[P.15.318] Genome-wide characterization and mining of microRNAs in the *Apis mellifera ligustica* larval gut reponse to Ascosphaera apis infection

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MicroRNAs(miRNAs) are endogenously expressed small, non-coding transcripts that play pivotal parts in insect-pathogen interactions. However, knowledge about roles of miRNAs play in honeybee larval responses to Ascosphaera apis is completely unknown. Here, we analyzed miRNAs expression profiling in *Apis mellifera ligustica* larval gut at different stages of A. apis infection via deep sequencing and bioinformatics. 694 known miRNAs and 46 novel miRNAs were identified. These miRNAs were 18-25 nucleotides in length, and the preference of the first base and base at each position of miRNAs with different length were obviously various. The Stem-loop RT-PCR results confirmed the true existence of novel miRNAs. Moreover, 34 markedly differentially expressed miRNAs(DEmiRNAs) in A. apis-stressed larvae guts of 4-, 5- and 6-day-old A. m. ligustica compared with normal larval guts. Further investigation demonstrated that DEmiRNA may participate in host responses to A. apis via regulating metabolism, cell processes and immune signaling pathways. Additionally, DEmiRNA-mRNA regulatory networks revealed complex regulation relationships between them, and miR-8440-y, ame-miR-927b, miR-429-y, miR-1638-x and novel-m0034-3p were located in the center, targeting the most mRNAs. Noteworthily, miR-8440-y was shared by all comparison groups, showing its vital part in host responses. Furthermore, miR-8440-y was showed to be involved in regulating various target genes associated with several immune pathways including ubiquitin-mediated proteolysis and Jak-STAT signaling pathway, further suggesting it may be momentous participant in host immunity defense against A. apis. In a nutshell, this is the first study on western honeybee miRNA response to A. apis infection from a genome-wide perspective, which demonstrates the expression pattern of miRNAs in A. m. ligustica larval gut was altered by A. apis invasion. DEmiRNAs may play an important role in the host stress responses including immune responses via negatively regulate target genes. These findings provide a foundation for elucidating the molecular mechanisms underlying the host responses, host-pathogen interactions and the miRNA-mediated cross-kingdom regulations during chalkbrood. miR-8440-y may be a promising candidate for future functional study and disease control.

[P.15.319] Combination of Varroa destructor and IAPV causes winter colony losses of *Apis mellifera* in China

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The combination of Varroa destructor and viruses is now considered the major cause of global colony losses of *Apis mellifera*. Large scale winter colony losses of *A. mellifera* were also frequently reported in China in recent years. To investigate the correlation of V. destructor infestation and virus infection and the subsequent effects on the overwintering performance of honeybee colonies in China, 45 colonies were kept without any Varroa treatment for two months to allow the propagation of V. destructor before overwintering in 2018. In October, the infestation rate of V. destructor was determined and the infection of 7 RNA viruses, namely Acute Bee Paralysis Virus (ABPV), Black Queen Cell Virus (BQCV), Chronic Bee Paralysis Virus (CBPV), Deformed Wing Virus (DWV), Israeli Acute Paralysis Virus (IAPV), Lake Sinai Virus (LSV) and Sacbrood Virus (SBV), was quantified. The overwintering performance of these colonies was then recorded. Of the 7 viruses, only DWV and IAPV were prevalent in the experimental colonies. The viral loads of DWV and IAPV were positively correlated with the infection rate of V. destructor, with correlation coefficient at 0.44 and 0.45, respectively. The correlation coefficient of DWV titer and V. destructor infection rate was higher in the colonies with infection rate lower than 10% (r=0.60), while that of IAPV and V. destructor was higher in the colonies with infection rate greater than 10% (r=0.62). Compared with colonies successfully overwintered, the 17 colonies died during overwintering had significantly higher IAPV titers and V. destructor infection rate, but not DWV titers, before overwintering. This study suggested that the synergistic effect of IAPV infection and V. destructor infestation was highly related to winter colony losses of *A. mellifera* in China and controlling the infection rate of V. destructor under threshold of 10% was important to reduce colony losses in winter.

[P.15.320] Genome-wide discovery and functional analysis of circular RNAs and their regulatory networks in western honeybee response to Nosema ceranae stress

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Circular RNAs (circRNAs) are newly discovered non-coding RNAs, which play key roles in various biological functions. Here, normal midguts of 7- and 10-day-old *Apis mellifera ligustica* workers (Am7CK, Am10CK) and N. ceranae-stressed midguts of 7- and 10-day-old A. m. ligustica workers (Am7T, Am10T) were analyzed using RNA-seq and bioinformatics. 10833 novel circRNAs derived from 2661 genes were identified from normal workers' midguts. The circRNA were mainly ranged from 15 nt to 1000 nt in length, and the number of circRNAs enriched in chromosome 1 in western honeybee was the largest. Additionally, the expressions of three circRNAs were confirmed using RT-PCR. 256 differently expressed circRNAs (DEcircRNAs) were identified in Am7CK vs Am10CK group. Their source genes were annotated to 32 GO terms and 35 pathways. The regulation networks indicated the DEcircRNAs might affect the midgut development as competitive endogenous RNAs (ceRNAs). Further investigation demonstrated 14 DEcircRNAs can target the same ame-miR-6001-3p. Besides, 61 (143) up-regulated and 107 (163) down-regulated circRNAs were predicted in Am7CK vs Am7T and Am10CK vs Am10T

groups. 44 and 121 source genes of the aforementioned DEcircRNAs were found to be associated with 27 and 35 categories including response to stimulus. Additionally, 25 and 33 source genes were enriched in 33 and 43 pathways, including several cellular immune pathways. Furthermore, the DEcircRNA-miRNA-mRNA networks showed 86 DEcircRNAs, 83 miRNAs and 215 mRNAs in Am7CK vs Am7T formed two networks; while 178 DEcircRNAs, 103 miRNAs and 305 mRNAs in Am10CK vs Am10T formed another two. These findings suggested DEcircRNA were likely to regulate gene expression in response to N. ceranae stress via absorbing corresponding miRNAs. Finally, 10 randomly selected DEcircRNA were examined by RT-qPCR, validating the reliability of our sequencing data. In conclusion, the number, type, structural property and expression pattern of circRNAs in the A. m. ligustica workers' midguts were comprehensively investigated followed by illustration of the roles of circRNAs involved in host responses to N. ceranae. Our results provide a valuable data resource for future study on bee circRNAs, but also novel insights into understanding the molecular mechanisms underlying western honeybee's responses to N. ceranae and host-pathogen interactions.

[P.15.321] Seasonal dynamics and phylogenetic analysis of viruses in two honeybee species, *Apis mellifera* and *Apis cerana*, in China

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Viruses are one of the main culprits in the current worldwide high levels of colony losses in managed *Apis mellifera* colonies. Contrast to the intensive studies on virus infection of *A. mellifera*, investigation on virus infection of *Apis cerana* is insufficient and the current status of virus inter-species transmission between the two species is also ambiguous. In this study, we collected adult bee samples from 361 colonies in 45 apiaries across six provinces of China for four consecutive seasons and detected the infection status of five common viruses, including Black Queen Cell Virus (BQCV), Chronic Bee Paralysis Virus (CBPV), Deformed Wing Virus (DWV), Israeli Acute Paralysis Virus (IAPV) and Sacbrood Virus (SBV) by quantitative realtime-PCR. In both A.mellifera and A.cerana, DWV was the most prevalent virus. The infection rates and viral loads of DWV, IAPV and BQCV on *A. mellifera* colonies were significantly higher than those of *A. cerana* colonies, while the result of SBV was just the opposite, suggesting different resistance of A.mellifera and A.cerana to virus infections. Seasonal variations of the four viruses were similar in the two honeybee species. Phylogenetic analysis revealed DWV, IAPV and BQCV freely disseminated across the honeybee species. SBV strains mostly clustered by the origin of honeybee species, with a few exceptions. Taking together with previous reports, this result indicated the species barrier of SBV infection has been overcome. This study provided the first report of seasonal dynamics of virus infection of honeybees in China. The cross-species infection of honeybee viruses poses new threats to both honeybee species. Further studies on virus infection of *A. cerana* can provide important reference for understanding the host-virus interactions in A.mellifera and may shed light on sustainable solutions to the losses of *A. cerana* and A.mellifera colonies caused by viruses.

[P.15.322] Investigating Host-Pathogen Interactions between *Apis mellifera* and Nosema ceranae

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Honey bees (*Apis mellifera*) are recognized as the world's most important insect pollinators for agriculture. However, many pathogens infect honey bees and pose a significant threat to honey bee health. One such pathogen is a microsporidian parasite known as Nosema ceranae. Nosema spores initially infect the honey bee gut, and the vegetative state rapidly proliferates, weakening the bee's immune system by depleting nutrients from the host. When daughter spores are fully grown in the gut cells, they rupture the cells, causing their release into the gut lumen where they can then be passed, via faeces, to other bees in the colony. As a result, the colony experiences a range of symptoms of nosemosis, the resulting disease, including dysentery, distended abdomen and weakness, all of which contribute to honey bee death.
Several preliminary experiments have been carried out to elucidate the effects of Nosema, although the molecular mechanisms of the disease are still poorly understood. Proteomic analyses looking at the infection dynamics of Nosema in honey bees have also been explored but its mechanism of action in the honey bee gut that results in disease has not yet been elucidated. We present preliminary studies using mass spectrometry-based proteomics aimed at exploring the mode of pathogenesis of Nosema in the honey bee gut. We have applied Protein Correlation Profiling - Stable Isotope Labelling of Amino Acids in Cell Culture (PCP-SILAC) to study how the host protein interactome changes in response to Nosema.

[P.15.323] Multi-stress effects on honey bee colonies

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High losses of honey bee colonies in recent decades are of great societal and economical concern and has been experienced as sign of vulnerability of agriculture, including the service of crop pollination, and of beekeeping. Varroa destructor infestation is acknowledged as an important cause of these losses and often suggested to act in concert with contributing stressors, such as low or monotonous food availability, infestation by Nosema spp. or exposure to insecticides. In several different field experiments, we studied the relative and interactive effects of V. destructor infestation and these stressors at field-realistic exposures on the performance and survival of honey bee colonies. In one study we found that ample food could not compensate negative effects by V. destructor on colony size or survival. In another study, colonies infested by V. destructor were 13% smaller in size and were 59.1 times more likely to die than colonies infested with low levels of V. destructor, but in contrast to the expectations no interactions with Nosema spp. or imidacloprid were found for colony size or survival. At individual level however, pollen foragers from colonies exposed to V. destructor in combination with imidacloprid flew less far (in a flight mill) compared to colonies exposed to a single stressor. Colonies as a superorganism may well be able to compensate at the colony level for negative effects of stressors on their individuals. In all of our experimental studies under field realistic exposure to stressors, V. destructor was by far the most lethal stressor for honeybee colonies.

[P.15.324] Microsporidian parasites Nosema apis & Nosema ceranae in honeybees – changes in time and species distribution

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In the last decades, beekeepers have observed increased colony mortality of honeybees (*A. mellifera* L.). One of the reasons for bee decline is believed to be the proliferation of diseases in honeybee colonies. For many decades it was considered that honeybees can only be infected by Nosema apis. However, studies made in Europe since 1996 shows that there are two species of the microsporidians causing the disease – N. apis and N. ceranae. The symptoms of infection by these two pathogens are very different, as are the virulence, spread and pathogenicity, which is why it is important to know the species distribution and changes in time.
As a result of acute nosema infection, the lifespan of infected bees is reduced, also metabolism can be affected, which results in both yield and pollination capacity. In combination with pesticides, this viral disease is even worse.
Five years ago, the EPILOBEE (2012-2014) study was conducted to map the spread of bee diseases in European member states. Then clinical symptoms of nosematosis were detected in spring in Estonia and Latvia. The aim of the present study is to find out which Nosema species infects honey bees in Estonia and Latvia, and is the disease still present in formerly positive apiaries.
Our study shows that in both countries in addition to N. apis, also N. ceranae is present and persists after the fifth year as well. N. apis was predominant in Estonia whilst N. ceranae was predominant in Latvia. In Estonia, the average number of spores per one worker bee was seven million and, in the neighboring country it was 2.18 million. The number of spores according to species will be discussed.
Nosematosis is very persistent. Even after the fifth year, about 75% of bee colonies are still infected. Combating infection is difficult because good medicine has not been developed so far. Further research is needed to clarify the reasons for different nosema species distribution in such a small area.

[P.15.325] Virus monitoring of Austrian honey bee colonies: Are virus titers correlating with symptoms in the field?

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At present, around 23 bee viruses are known, which can weaken or even kill an infected honey bee colony. However, representative data about the prevalence of most bee viruses in Austrian colonies and the size of their impact onto colony losses are missing. In the frame of the project “Zukunft Biene 2” we cooperate with 200 Austrian beekeepers to determine the prevalence of eight bee viruses in Austrian apiaries (ABPV, BQCV, CBPV, DWV type A and B, IAPV, KBV, SBV). Each beekeeper selected one of their apiaries for sampling. In three consecutive years (2018-2020), they sample five of their colonies in September and send us living bees in queen cages, as well as a filled-in questionnaire (beekeeping practices, colonies’ health status, visible virus symptoms). Samples are frozen immediately upon arrival (< -18°C), pooled according to each apiary and analysed quantitatively for the eight viruses by RT-qPCR. In the first year of the study, 198 of the participants sent in the samples (September 2018). The following three symptoms were observed frequently by the participants in at least one colony: black shiny bees (33 % of the participants), varroa mites on bees (18 %) and bees with deformed wings (10 %). Six of the eight viruses were detected in at least one sample: ABPV, BQCV, DWV type B (VDV-1) and SBV were highly abundant, whereas CBPV and DWV type A were found less frequently. IAPV and KBV were not detected. The virus data will be related to the occurrence of visible virus symptoms and the winter losses of the particular apiaries in the season 2018/2019.

[P.15.326] A new report of multiple honeybee viruses in the Invasive hornet, Vespa velutina

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Honeybees are infected by viruses that have a broad range of hosts and harmful to other insect pollinators, including *Bumblebee*s and other wild bees. In addition to honeybees, Asian hornet, Vespa velutina, colonies are managed and harvested as delicacies in Yunnan, Southwest China. Hornet keepers usually fed the hornets by keeping a honeybees apiary near the hornet colonies. We are wondering whether honeybee viruses are being transmitted to V. velutina through preying on and consuming infected honeybee and if the viruses are undergoing genetic adaptations within the hornets. Seven common honeybees viruses were screened and self-replicating Deformed wing virus (DWV) and Israeli acute paralysis virus (IAPV) were found in both the *A. mellifera* and V. velutina individuals from the same site. Interestingly, no viruses were detected in Asian hornets sampled from a site without *A. mellifera* colonies. The highest DWV-A copies were recorded as 1E11 of the whole symptomatic hornet; by adding the factor that Asian hornet is bigger than honeybees, the titer was similar to that reported in *A. mellifera* with deformed wings. Deformed wings symptoms and virus infection intensities in V. velutina were surprisingly similar to that in *A. mellifera*. Phylogenetic analysis of virus RdRp genes suggests the identified viruses from V. velutina are still grouping with that identified in the honeybees, suggesting few or no genetic changes were needed for the viruses to infect the hornets. This study further supported the idea that some viruses, i.e. DWV and IAPV, found in honeybees should be universal to other hymenopteran insects. Honeybees could be acted as a good virus reservoir in the same environment and affect other hymenopterans that could have naturally prevented virus diseases.

[P.15.327] Enhancement of MAPK signaling pathway after deformed wing virus (DWV)-infected honey bee larvae

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Deformed wing virus (DWV) is high prevalence in the honey bees (*Apis mellifera*) populations. It could infect honey bees via vertical and horizontal transmissions. However, there is little known about the impacts of DWV on honey bee larvae and consequence of deformed wing symptom. To evaluate the pathogenesis of DWV in the larval stage of honey bees, the artificial rearing honey bee larvae were infected with DWV (1.55x1010 copies/per larva). The survival rate of infected larvae decreased significantly from 6 days (pupal stage) post infection (d.p.i.). The detection of viral replication increased from 2 d.p.i. and the highest viral copies was detected from 4 to 6 d.p.i. Moreover, the DWV copies were increasing continuously in the pupal stage. Based on the results, the DWV-infected and control groups were than subjected to transcriptomic analysis at 4 d.p.i. The results of the transcriptomic analysis showed that total 1195 differential expression genes (DEGs) were identified. Of these DEGs, 488 were down-regulated genes and 707 were up-regulated genes. The result of GO(Gene Ontology) showed 564 DEGs (65.1%) were categorized into molecular function, cellular component, biological process and high percentage of DEGs involved in cellular and metabolic processes. Total 365 (217 up-regulated genes and 148 down-regulated genes) were annotated by KO (KEGG Orthology) pathway mapping and highly hit to metabolic pathways, Biosynthesis of secondary metabolites and MAPK signaling pathway. Besides, six highly expressed genes, including Gld, Abaecin, Hymenoptaecin, Apidaecin, AmPPO and CelD were also detected. Among these genes, total 4 genes are immuno-related genes, these result indicated that the enhancement of the MAPK signaling pathway-related genes and immuno-related genes may play important role in infected honey bee larvae against DWV infection. Together, the predicted DEGs in the MAPK signaling pathway and immune-related pathway will be selected for further validations. The aforementioned information would provide the new insight to the deformed wing of honey bee after DWV infection.

[P.15.328] The invasive hornet Vespa velutina carries both honey bee viruses and new viruses

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From its first introduction in 2004 in France the Asian yellow-legged hornet Vespa velutina nigrithorax is now largely spreading into Europe, by lack of efficient control methods. In this context, looking for natural hornet pathogens could be useful to develop biological control agents against this invasive species. Therefore, we investigated all RNA viral sequences detected from hornets samples caught in Southern France, in order to characterize any virus from asymptomatic or symptomatic (deformed winged) hornets differentiating various dissected parts (brain, muscle and abdomen).

Among almost twenty viruses detected in three RNA sequenced hornets, Deformed wing virus B (DWV-B) was shown to be predominant in all samples, and much higher in the muscle from symptomatic individual, suggesting a putative cause of the deformed wings symptom. Interestingly, two new viruses closely related to Acyrthosiphon pisum virus (APV, unclassified ssRNA+ virus) and Himetobi P virus (HPV, Triatovirus, Dicistroviridae) were detected in brain and muscle that may correspond to the circulation and multiplication forms of those viruses in the hornet.

From a multiplex PCR method, two bee viruses, Acute bee paralysis virus (ABPV) and Black queen cell virus (BQCV) were detected in brain, muscle or other dissected parts. Other viruses known to infect the honey bee were also identified from the hornet intestine transcriptome such as Aphid lethal paralysis virus (ALPV), Bee Macula-like virus (BMLV), and Mokuvirus.

Our study underlines the urgent need to study the host range of these original new viruses we described in hornets to evaluate if they can represent a new threat for the honey bees or a hope for the biocontrol of V. velutina.

[P.15.329] Pathogen prevalence and risk factors for winter loss of honey bees in Southwestern Quebec

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Winter mortality of bees represents a major source of economic losses for the beekeeping industry. Pathogens, management practices and environmental factors are thought to be involved in this phenomenon, but their exact roles and interactions between one another are still poorly understood. This pilot project aims to estimate the prevalence of the main pathogens of the honey bee and determine the impact of these risk factors on overwintering loss in Southwestern Quebec.
The pathogens Varroa destructor (detected by alcohol wash), *Paenibacillus larvae*, Melissococcus plutonius, Nosema spp., Acarapis woodi and Apocephalus borealis were investigated on adult bees in 242 colonies of 31 apiaries of Pontiac and Montérégie areas in the province of Quebec in August 2017. Colony mortality (<4 frames of bees) was registered in May 2018. The prevalence of each agent was estimated at the colony and apiary levels. Varroa was the most common pathogen, found in 48% of colonies and 93% of apiaries. However, only 16% of colonies in 49% of apiaries had significant infestation rates of at least 1 mite per 100 bees. Nosema spp. and M. plutonius were respectively detected in 40% and 21% of colonies. A. borealis was detected in one colony. A. woodi and P. larvae were not detected. Overall colony mortality was 31%, whereas intra-apiary mortality ranged from 0 to 100% with a mean of 37% and a median of 30%. The impact of pathogens, clinical signs of disease, environmental factors and management practices on colony mortality over the subsequent winter, based on logistic and negative binomial regression models, will be presented.
The preliminary data shows that Varroa remains the most frequently encountered pathogen in apiaries in southwestern Quebec. Given the recognized impact of Varroa on colony health, it is essential to establish a strategy for effective monitoring and management of infestation levels. The findings of this study will provide useful information for the implementation of targeted and effective preventive medicine programs.

[P.15.330] Transcriptomic investigation of immune responses of the *Apis cerana cerana* larval gut infected by Ascosphaera apis

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Chalkbrood is representative fungal disease for honeybee. The objective of this work was to reveal immune responses of *Apis cerana* cerana larval gut following Ascosphaera apis invasion. Our group previously validated drone and worker of A. c. cerana can be infected by A. apis using a combination of morphological and molecular approaches. Combining a previously assembled transcriptome of A. c. cerana larval gut and the obtained high-throughput sequencing data in the present study, 6152 differentially expressed genes (DEGs) were clustered into eight profiles including three significant profiles and three down-regulated profiles following trend analysis. GO term analysis suggested DEGs within significant up-regulated and down-regulated clusters were enriched in 46 and 38 functional groups, respectively. KEGG pathway enrichment analysis indicated a majority of DEGs were involved in ribosome, carbon metabolism, biosynthesis of amino acids, and oxidative phosphorylation. In addition, 142 and 14 DEGs were respectively annotated in the cellular immune- and humoral immune-related pathways. Further investigation notified that DEGs up-regulated in cellular immune and humoral immune pathways outnumbered those down-regulated. Moreover, immune responses of A. c. cerana and *Apis mellifera ligustica* larvae were compared and investigated to decipher resistance of eastern honeybee larvae to A. apis. These results demonstrated that an overwhelming number of genes involved in immunity-related pathways were activated by A. apis. In summary, for the first time, a comprehensive transcriptomic investigation of immune responses of A. c. cerana larval gut to A. apis infection was conducted based on RNA sequencing and bioinformatics. Our results not only provide valuable information for revealing the molecular mechanisms underlying immune responses of A. c. cerana to A. apis and host-pathogen interactions during chalkbrood, but also lay a foundation for illustrating the key factors responsible for the A. apis-resistance difference between eastern honeybee and western honeybee.

[P.15.331] Insights into the Molecular basis of Host-Parasite Interactions between Honey bee and Nosema apis

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Honey bees (*Apis mellifera*) are key pollinators of agricultural and environmental importance but their pollination services are under threat following global declines in a number of managed and wild bee populations. Parasites are known key contributors to declining bee health and we used a globally widespread fungal pathogen, Nosema apis, to unravel the molecular interplay that determines both parasite virulence and honey bee resistance. To further, understand the drivers that determine Nosema virulence, we passaged Nosema through nine different host bee genotypes for a total of three cycles. We expected that our experimental procedure would select different sub-populations (or strains) from our original and genetically diverse Nosema pool and confirmed this idea through genetic and phenotypic follow-up experiments. In the last step, we sequenced the genomes of all nine serial passaged Nosema samples to link genetic variation

with differences in Nosema virulence. We also investigated the mechanisms of host resistance against parasite virulence as previous findings had indicated that honey bees produce multiple molecules which independently kill Nosema spores. Here we provide empirical evidence that the protein chitinase is involved in the killing of Nosema spores. Our results provide not only very detailed insights into the complex molecular interactions between a honey bee parasite and its host but offer novel opportunities to manage the diseases in the future; for example, through targeted breeding programs or by developing novel medication for the treatment of infections.

[P.15.332] Factors influencing honey bee colony losses throughout Mexico during 2017

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This study was conducted to identify environmental risk factors, diseases and agrochemicals, that may be associated to losses of honeybee colonies or to a decrease of hive productivity in Mexico. A survey was applied to 1,860 beekeepers and samples of adult bees, brood and beeswax, were randomly collected from 368 apiaries. Beekeepers and colonies from Mexico's five apicultural regions (Yucatan Peninsula, Pacific, High plateau, Gulf and North) were included in the study. The survey allowed to establish the beekeeper's profile, determine apiary management practices, identify diseases and treatments used, as well as to record the percentage of annual colony losses and their possible causes. The mean number of years of beekeeping experience among those interviewed was 18, and 97% of them mange bees mainly for honey production. Beekeepers reported colony losses ranging from 0 to 10% and a mean decrease in honey yields of 20%, being climate change the most common cause named for these problems. Analyses of the samples identified pathogens, particularly Varroa destructor, as the main disease agent (80% frequency with a mean infestation level of 3.5%). Nosema spp. was the second most frequent pathogen found (36.9%), whereas viruses such as the deformed wing virus and sac brood bee virus were also highly present (31.3 and 21.1%, respectively). Agrochemicals in beeswax were detected in 8% of the samples, mainly in those from the Gulf and North regions, where 22 and 20% of the samples, respectively, had pesticide residues (the most common being chlorpyrifos with concentrations of up to 1.1 ppm). American foulbrood, Kashmir bee virus and chronic bee paralysis virus were not detected. It is concluded that parasitic diseases, viral diseases and climate change, could be the main risk factors associated to colony losses and to a downward trend in apicultural productivity of Mexico's beekeeping industry.

[P.15.333] Impact of Vespa velutina on honeybees and other pollinators

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The spread of invasive alien species and the decline of pollinators are two main issues that must be handled for the conservation of nature and biodiversity. Pollinators are a group of animals of primary importance - mainly insects like honey bees, wild bees, hover flies, butterflies, moths and beetles - which contribute to pollinate crops and wild plants and that are facing a serious decline in occurrence and diversity in the last decades. The Asian yellow-legged hornet (Vespa velutina) is an invasive alien species that represents a serious threat to European pollinators. It is known to be a fierce predator of honey bees, but can also hunt in particular wild pollinators and overthrow native wasps. Beside to the control activities developed by the LIFE STOPVESPA project against this invasive hornet, the impact of V. velutina on pollinators has been evaluated throughout the analysis of the status and trends of wild pollinators and wasps communities in Liguria region, Italy. Different study areas were selected at various level of density of the Asian yellow-legged hornet. Pollinating insects and wasp communities were periodically sampled using two types of traps (baited bottle trap and pan trap) in different seasonal periods. In total, 11,682 samples, classified in 40 genera and 32 species were collected. We found 25 species of wild bees not previously reported in Liguria region and one new species for Italy (Andrena asperrima). V. velutina nest density clearly affect insect communities (significant drop in European hornet, wild bees and diurnal butterflies abundances) besides to the effect of climatic factors and landscape features.

[P.15.334] Long-term Monitoring the *Apis cerana* Sacbrood Virus Prevalence Rate in Northern Taiwan

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Since 2016, *Apis cerana* sacbrood virus (AcSBV) have been recorded in Taiwan and causes serious loss and epizootic in *Apis cerana*. Most of the *A. cerana* populations are reared in Northern Taiwan and our previous spatial investigations revealed that the highest AcSBV prevalence rate was also detected in Northern Taiwan. Herein, we performed the two yaers long-term survey of AcSBV prevalence in the populations of A.cerana at Northern Taiwan from January 2017 to 2018. From our survey, the average prevalence rates of AcSBV were about 76.5% and 48.5% in A. cerna and *A. mellifera*, respectively in 2017, while decreased to 37.7% and 20.4% in 2018. The average prevalence rate in A.cerana were general higher than those in *A. mellifera*, and the epidemic has been ebbing at 2018. Interestingly, the AcSBV prevalence rate from the apiaries of two species gradually converted to a similar trend especially in the last four months. In our observation, AcSBV can cause the typical symptom when the prevalent rate higher than 62.5% but it may loss the advantage of virulence in *A. mellifera*.It is worth noting those co-cultured apiaries would have higher prevalence rate than *A. cerana* or *A. mellifera* single-cultured apiaries. The possibility of AcSBV interactive transmission or among *A. cerana* and *A. mellifera* was observed. Our data provided a long-term monitoring data of this epidemic and that may be a chance to see the prediction of AcSBV for the future. Regarding the symptom caused by AcSBV is quite different between these two honey bee species; we herein propose that *A. mellifera* may be as a guard honey bee species to monitor the AcSBV infection in the population of A.cerana.

[P.15.335] Detection of deformed wing virus and black queen cell virus in parasitic mites, Varroa destructor, from Iranian honey bee (*Apis mellifera*) colonies

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The parasitic mite Varroa destructor and the viruses it transmits to honey bees (*Apis mellifera*), are often mentioned as culprits of colony losses. Therefore, surveys are conducted in different countries to determine the presence and distribution of honey bee viruses. In Iran, the information about honey bee viruses is scarce and limited to their presence in honey bees, but have never been reported from V. destructor. We conducted a study for detecting five viruses in samples of V. destructor mites that were collected from 12 apiaries located in the Iranian provinces of Alborz, Ardabil and Gilan. RT-PCR was used to diagnose the viruses, including deformed wing virus (DWV), black queen cell virus (BQCV), acute bee paralysis virus (ABPV), Israeli acute paralysis virus (IAPV) and Kashmir bee virus (KBV). Several mite samples resulted positive for DWV and BQCV, but all were negative for ABPV, IAPV and KBV. Minus DWV RNA strands were detected with tagged RT-PCR. This is the first report of honey bee viruses found in V. destructor mites collected in Iran, the first to report BQCV in Iranian honey bee colonies and the first to demonstrate that DWV replicates in V. destructor mites from Iran.

[P.15.336] Prevalence and Risk factors associated with Varroa mite infestation and its Effect on honeybee Colonies (*Apis mellifera jementica*) in Northern Ethiopia

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The ecto-parasitic mite, Varroa mite, has become the largest threat to apiculture and honey bee health worldwide. However, the effect of the mite on the honeybee colony strength has not yet studied in Ethiopia. Hence, this study was conducted in honeybee colonies in Tigray Region, Northern Ethiopia between September 2014 and June 2015 to determine the prevalence, infestation level, identify associated risk factors and association of Varroa mites with colony strength. A total of 87 apiary sites and 384 honeybee colonies were randomly sampled and inspected internally and externally for the presence Varroa and colony strength. The major risk factors for Varroa mite in honeybee colonies were agro-ecology ($p<0.01$), comb age ($p<0.05$), colony strength ($p<0.05$) and seasons ($p<0.05$).The overall infestation rate of Varroa mite was 1.8%.The average number of Varroa mites recorded during honey flow season was significantly higher than during the dearth period ($p<0.01$). The number of Varroa mite in honeybee colonies was not significantly correlated with honeybee colony strength ($p>0.05$). At this stage, Varroa mite have no a significant negative effect on honeybee colony strength. Long term monitoring and mite threshold level determination is recommended to be studied.

[P.15.337] Varroa destructor mites irritate honey bees and make them sting faster

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Honey bees (*Apis mellifera*) protect their nests and food stores from intruders by stinging. The stinging behavior of honey bees could be affected by a number of stressors, which could reduce or increase their ability to defend their nests. However, there are no studies on how particular stressors, like parasites, affect this behavior. Among the parasites affecting honey bees, the most damaging is the mite Varroa destructor. V. destructor is an external parasite that feeds on honey bee hemolymph and fat body tissue, causes serious health problems and increases bee mortality. This study was conducted to determine if V. destructor could affect the stinging responses of individual honey bees in a controlled environment. Groups of 30 bees of the same age were subjected to one of the following treatments: 1) control bees that were fed sugar syrup and kept in cages in an incubator until tested, 2) bees artificially infested with two mites/bee, kept under the same conditions as the control bees. When the bees were seven days old, they were exposed to an electric stimulus of 0.5 mA, and the time they took to sting a leather patch was recorded under laboratory conditions. The experiment was repeated three times. Bees exposed to Varroa parasitism stung significantly faster (2.58 ± 0.29 s) than the control bees (3.63 ± 0.52 s; $P<0.005$). It is concluded that Varroa increases the irritability of the bees and reduces their stinging response time. The implications of these results on nest defense will be discussed.

[P.15.338] The effect of the infection by the ectoparasite Varroa destructor on the physiological performance of *Apis mellifera*

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Varroa destructor is the responsible for the more important disease in honeybees and beekeeping. The effect inside the hive like the health and production capacity is well known. It is well documented too that Varroa is a vector of pathogens and is responsible for malformations in the brood. But there is a lack of information about what happen with the physiology response in the bee when is parasited by Varroa. Within this context, the aim of this study is to test how is the physiology response and the survival of the honeybees when the ectoparasite Varroa destructor is present. We conducted a laboratory essays using 32°C as acclimation temperature with a humidity of 65% for 14 days (the last 7 as brood phase and 7 days as young workers). When the acclimation ended, we selected 30 honey bees and designed three treatment groups of bees: a) control bees (without mites), b) treated bees with 1 mite and c) treated bees with 2 mites. After one hour of direct and effective parasitization, we measured individually the metabolic rate during 3 hours of 10 bees per mite treatment in a metabolic chamber. We observed that the metabolic rate was significate higher when the mite number on the bees increased. The metabolic rate increased by 50%, compared between treatments.
In a second series of experiments, we measured the survived of each bee individually in the same treatment (0, 1 and 2 mites) for 8 days. We used 40 bees per treatment. We could see that the survival of the bees is less than 20% comparing between 0, 1 or 2 Varroas.
We conclude that the performance of bees is affected negatively by the environmental increased temperatures and parasite load under laboratory conditions.

[P.15.339] Nosema ceranae, the most common microsporidium infecting *Apis mellifera* in the main beekeeping regions of China since at least 2005

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The aim of this study was to determine the distribution of Nosema spp. in *Apis mellifera* using historical worker bee samples collected between 2005 and 2010 in China. Out of 292 samples initially analysed by microscopy, 69 were Nosema spore positive. These samples were subjected to multiplex PCR amplifications with primers corresponding to 16S rRNA specific sequences of N. ceranae and N. apis. The results demonstrated that N. apis was detected in only one sample collected in 2008 in Shandong province, whereas N. ceranae was detected in 68 samples, which indicates that N. ceranae is the most common Nosema species infecting *A. mellifera* in China since at least 2005.

[P.15.340] Are Drones the Missing Link in Colony Losses of Honey Bees?

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Declines in native insect pollinator populations and substantial losses in managed honey bees (*Apis mellifera*) have been reported on a global scale and become a widespread concern because of the importance of these insects for human food production and ecosystem stability. Several factors have been investigated as causes, such as parasites and pathogens, exposure to agricultural pesticides, habitat loss and/or climate change. More recently, a combination of potential factors have been studied as possible causes of declining pollinator health. Here we focussed on the involvement of drones in pollinator losses. Pesticide as well as parasite infections affect drones differently than workers and the combination of these stressors has devastating effects on drone survival. We found that the vast majority of males did not survive to sexual maturity after exposure to very low levels of the neonicotinoid, Thiamethoxam and also Nosema apis infections. This does not only reduce the reproductive success of individual colonies, but can also impact gene flow and genetic diversity at the population level, which are both known as key components of honey bee health.

[P.15.341] Actual situation of Aethina tumida (Coleoptera: Nitidulidae) in Africanized honeybee colonies in Costa Rica

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During the last years, the small hive beetle, Aethina tumida, has been found in Africanized honeybee colonies in several countries of Central America. It was detected in El Salvador in 2013 and Nicaragua in March 2014. In Costa Rica, A. tumida was confirmed in Africanized honeybee colonies in August 2015. Only adults were detected in a sentinel apiary monitored by the National Service of Animal Health (SENASA). For its economic importance and biological significance, this study was conducted to monitor the distribution and report the actual situation of the SHB in Costa Rica. A total of 77 apiaries located in five provinces were sampled from August 2015 to October 2018. Colonies were monitored visually by examining all individual frames, hive covers, and bottom boards for each colony. Furthermore,

two Cutts Beetle Blaster® traps were placed per colony, during eight to 15 days, with 25 ml of vegetable oil. The content of the traps was examined for beetles at the Bee Pathology Lab of the Tropical Beekeeping Research Center (CINAT). Furthermore, 10 wild colonies (swarms) located in trees or houses in urban areas of the Central Valley were also analyzed. A survey recorded no adult SHB in a total of 476 colonies sampled suggesting that it is not well established after its introduction in 2015. Nevertheless, three adult specimens were unexpectedly found in a wild honeybee colony located in a house in an urban area of Heredia-Central Valley in September 2018. The beetles were analyzed and preliminary determined to be Aethina tumida using definitive morphometric characteristics. SENASA confirmed by PCR analysis that the beetles were indeed A. tumida. The first report of A. tumida in the Central Valley of Costa Rica is provided to alert beekeepers and regulators to be vigilant to prevent diffusion of this invasive pest throughout country. Such information is also important because a number of recent studies suggest that the beetle may be able to exploit colonies of other social bees as alternative host. It should be mentioned that in Costa Rica there are more than 55 species of stingless bees.

[P.15.342] Reasons for high winter losses; Based on studies performed in humid and dry regions of Iran, North and South

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Usually high winter losses of bee colonies can be caused by Varroa mite infestation, diseases, protein deficiency, starvation, queen failure, and poor mating. This study was performed in the humid and rainforest region of North Iran and in dry region of South Iran, Persian Gulf.
In North of Iran the reasons for high winter losses were, an increase in the level of Varroa mites caused by non-stop brood production; protein deficiency caused by long period rain; high number of bees per region – which is one of the causes of epidemic diseases and shortage of pollen/nectar; a high level of moisture, and supercedure of queens due to poor mating during the long period of rain.
In South of Iran the reasons for high winter losses were due to field bees being attacked by wasps, hornets and birds, which results in the low flow of pollen in the hives and low level of brood production; also due to the lack of rain and poor vegetation, during August and September, colonies become malnutrition, brood productions reduces significantly, and bees reduce the number of drone bees in the colonies and the remaining drones do not produce enough semen, as a result, hives experience a large number of queen failure. These challenges are prevalent among countries in Persian Gulf regions due to the long hot season.
In North of Iran, to prevent winter losses, Varroa mite should be controlled control, proper insulation and ventilation within the hives, and pollen patties are required during September.
In South of Iran, it is difficult to control wasps, hornets and birds. Also, providing pollen patties is not cost effective, due to the long period of dry season. Therefore it is recommended to transport the bees to the higher elevations during August and September.
In this presentation the causing factors of winter losses in 2 opposite extreme conditions, humid and dry, will be discussed in further details.

[P.15.343] Welfare of managed honey bees - a holistic approach to bee health

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Honey bee (*Apis mellifera*) colonies are considered “superorganisms” as individual bees do not display the complete behavioural and ecological range of the species. The colony acts as a functional entity, based on division of labour. These social insects are in a close relationship with the environment. As a reared animal, *A. mellifera* has spread far beyond its areas of origin, now living in all inhabited continents. This dispersal made them confront novel stressors, like unsuitable environments and management practices or new pathogens and pests. The severity of these factors extensively eradicated the wild honey bee population in many areas of the world, where the species survives only thanks to domestication.
The high resilience of honey bee colonies against disturbances probably led beekeepers to overlook the colony welfare for a long time. Nevertheless, increasing importance is now attributed to honey bee health, also for its economic impact on the honey crop and other productions. We propose to extend the concept of bee health to bee welfare. Following the approach established by OIE for vertebrate welfare, we introduce a new concept for managed honey bee colonies including pest and pathogens, nutrition, environmental stressors like modern agriculture and global warming as well as propositions for better practices for maintaining bee health.

[P.15.344] Mass measurement of single Varroa destructor: what that can tell us more

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Measurement of live animals is always a problem. They move causing instability of measurement. Or they are too light to be measured accurately. Either way, one has to follow what methods are available, and if not, develop a new one. Everything stated above explains why there are not weighing results of mass for single varroa ontogenetic stages of the adult female mite published in scientific literature. The searched literature review showed that mite had just two-dimensional data of their size.
We measure mass or weight of Varroa destructor mite from worker brood of *Apis mellifera* carnica at the time when adult bee emerged from brood. We weighing both animal species under similar conditions, real-time, under ISO 17 025 accredited laboratory and guided with requirements given in Metrological guidelines and Good Weighing Practice. We use just ten mites and ten bees. Results give us awareness on ratio and relationship of body weights comparing possible impact that parasite with his body weight can have on western honey bee
We try to understand and point evolutionary and ecological pest- host relationship of varroa load weight, not just see varroa as a vector for bee disease. The physical influence that newly added weight (average value: 0,43 mg) can have on the single bee as well as for social organism is something that we want to understand more. There are lots of associations that can have an influence on future research. We are just giving pioneering data of first varroa weighing results. Much more modelling and research scenarios can be created in future concerning the newly recognized physical value and possible pathophysiological effects in the hive.

[P.15.345] Molecular Detection of Nosema species from *Apis mellifera* from Northern India

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The aim of the study was to detect the infection level of honey bees with Nosema apis and/or Nosema ceranae using microscopic and molecular analysis from *Apis mellifera* from different beekeeping regions of Northern India. A detailed survey was conducted and 90 apiaries were chosen at random from these locations. Infection level was determined both by microscope and Multiplex-PCR and data were analyzed using bioinformatics tools and phylogenetic analysis. Result showed that N. ceranae was the major species infecting *A. mellifera* colonies in northern India. Only in six apiaries N. apis has been found coinfected with N. ceranae. Nosema spores were found to be in 12.50 % of total samples colonies, while 46 % of the samples evaluated by PCR were found to be positive for N. ceranae, with the highest prevalence Saharanpur region of Uttar Pradesh Provenance. This is the first comprehensive report about the Nosema detection, contamination level and distribution pattern in northern India

[P.15.346] Varroa mite infestation: a threat to nigerian beekeeping industry

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This presentation is a field report from our experience in Nigerian Beekeeping industry using Enugu and Ebonyi States as case study. These two states are located in the South East geopolitical zone of Nigeria. This study reveals the presence of Varroa mite in some inspected Apiaries within the study areas and also explains some deductions from eight years of field experiences with Varroa mite.
Varroa mite is an ectoparasitic mite which attacks all lifecycle stages of many honey bees species, including the *Apis mellifera* and all its subspecies. There are currently different genotypes of the varroa mites. At the South Africa Apimondia meeting, Dr. Anderson reported that over twenty genotypes of newly-named Varroa destructor and newly-defined Varroa jacobsoni now exist. The presence of varroa mites has been reported in most parts of the world.
In the month of March, 2010, we identified few of this mite in some of our honey bee colonies. This infestation was ignored because the colonies were still performing well in terms of population (strength), honey yield and usual hive activities. The assumption inferred from this scenario was that our bees are resistance to Varroa. From 2012 to 2016, Colony inspection records revealed that 70% of fifty colonies under study had varroa mite at different levels, the honey yield reduced and population of the bees reduced drastically.
In 2018, Dr. Usman Dukku, a researcher from Abubakar Tafawa Balewo University Bauchi sampled ten out of thirty colonies from our Apiaries. Soap wash method was used to detach varroa from adult worker honey bees and all the ten colonies were varroa positive. Also findings from his research work on Varroa mite in Bauchi State (Northern Nigeria) shows that from the fourteen honey bee colonies sampled in two different locations, twelve colonies recorded varroa positive (Usman and Sandra, 2017).
Therefore, by these findings, we strongly recommend an urgent intervention into research on honey bee health in Nigeria Beekeeping field to identify the genotype of varroa mite in the country and its prevalence. The research findings will help develop appropriate organic and safe measures to fight varroosis in Nigeria.

[P.15.347] A nine year survey of honey bee viruses in argentina. 2009-2017

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Honey bee (*Apis mellifera*) is one of the most important insect pollinators that ensure crop pollination. However in the last years, bees have been experiencing considerable losses worldwide. Several factors have been proposed to explain this losses, such us beekeeping practice, environmental conditions, use of agrochemical and relationship with pathogens, viruses among them. The aim of this study was to analyze the prevalence of bee viruses during a nine year survey (2009-2017) in Argentina. The sampling was designed according to a division of beekeeping regions of our country proposed by CFI-INTA (Consejo Federal de Inversiones – Instituto Nacional de Tecnología Agropecuaria). The percentage of samples from each region was calculated according to the numbers of hives distributed in each one and their relation with the total number hive through Argentina. Every year we take 80 samples from the five regions of country as show: Central region (n= 53), North East (n= 11), Patagonia (n=7), New Cuyo (n= 5) and North West (n=4). One colony per apiary was sampled. In total we study 720 samples. We use a multiplex PCR that can detect IAPV, DWB, SBV, ABPV, BQCV, CBPV and KBV. KBV was the only virus that was not found in the survey. DWV (28%), IAPV (23%) and SBV (21%) were the most prevalent viruses. Most positive hives was detected in 2010 with 96.3% of them at least with one virus. Eighty nine samples (12.4%) harbored more than one virus. Among then 66 samples where co-infected with 2 viruses, 12 samples with three virus and 11 samples with four viruses. As it happened almost all around the world, DWV was most prevalent virus in hives of Argentina (62 samples). However IAPV was the second most prevalent virus (32 samples). This last result is not so common and only exist one report in Spain in 2006-2007 with similar results that our study. The high prevalence of IAPV all around Argentina and the new reported cases by Colony Collapse Disorder turn on an alert and push us to develop different alternatives to the control of these viruses in Argentine apiaries.

[P.15.348] Does the recently discovered phorid fly in Saudi Arabia explain the unexplained losses of *Apis mellifera* colonies in other regions?

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Recently a phorid parasitoid infesting *Apis mellifera* carnica colonies has been discovered in Saudi Arabia. The case was reported to the Journal of Apicultural Research https:// doi.org/10.1080/00218839.2018.1466760. The study indicated three assumptions for its incidence: (a) it probably entered as invasive parasitoid through imported bumble bee shipments from the Netherland. (b) It might have been introduced in A. m. colonies imported from Egypt. (c) It may be present in the country during the past years infecting other hosts rather than honey bees, and never has been diagnosed.
Phoridae are small, poorly –identified flies' world- wide. The zombie fly, previously documented by infesting A. m. colonies, only in the USA, and the currently discovered hump-backed fly, that is not fully identified in Saudi Arabia, may explain the reported unexplained losses of honey bee colonies in Saudi Arabia and other countries. Moreover, it is proved to harbor to A. m. colonies Nosema ceranae and deformed wing virus. Infested honey bees abandon their hives during night leading to CCD. The study highlights on the importance of world-wide collaborative research work to evaluate the status and possibly to identify new species or mimics of zombie flies.

[P.15.349] Honey bee colonies of African maternal lineage are less susceptible to Varroa destructor infestations and to viral infections than colonies of European maternal lineage

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This study was conducted to test the hypothesis that honey bees of African maternal lineage are less susceptible to Varroa destructor infestations and to viral infections than bees of European maternal lineage. A total of 233 colonies from Jalisco, Mexico, were evaluated for adult bee populations (frames covered by bees), and V. destructor infestation levels in adults (alcohol washes) and brood (inspection of worker brood cells). Bees from 72 colonies were also tested for presence of deformed wing virus (DWV), black queen cell virus (BQCV), Israeli acute paralysis virus (IAPV) and chronic bee paralysis virus (CBPV) using RT-PCR. Additionally, the colonies were analyzed and classified by haplotype, depending on whether their workers had African or European mitochondrial DNA. Of the 233 colonies, 131 and 130 had African and European mt. DNA, respectively. The data were analyzed with non-parametric tests. Nearly 90% of the colonies were infested with V. destructor with no bias by genotype. There were no differences for adult bee populations between colonies of the two types ($P> 0.05$), but the colonies with African maternal lineage were significantly less infested with V. destructor mites (4.2±0.04 and 3.4±0.03% for adults and brood) than the colonies with European maternal lineage (5.2±0.03 and 5.9±0.06% for adults and brood). For viruses, only DWV and BQCV

were detected, and again, a significantly lower proportion of colonies with African maternal lineage were infected with DWV (22.9%) compared to colonies with European maternal lineage (48.6%). Furthermore, significantly fewer colonies with African maternal lineage were infected with BQCV (42.9%) than colonies with European maternal lineage (70.3%). Our data support the hypothesis that honey bee colonies of African maternal lineage are less susceptible to V. destructor infestations and to viral infections than colonies of European maternal lineage.

[P.15.350] Winter colony losses 2015/16 in Austria: are there correlations with pests, pathogens and pesticide residuals?

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Winter losses of honey bee colonies occur every year, but the extent differs between years and regions. A main problem in identifying reasons for winter colony losses is the lack of adequate sampling matrices at the time of discovery of dead colonies. Using a posthoc approach, we aimed to identify critical parameters and risk factors for winter losses. We selected 188 apiaries representative for Austria and inspected these in autumn 2015 and spring 2016. From up to ten colonies of these apiaries bees and beebread were sampled in autumn, immediately frozen and stored until spring. After evaluating survival or death of the sampled colonies in spring, all 60 dead and 150 randomly selected surviving colonies were chosen for detailed analysis (bee samples: ABPV, CBPV, DWV, Nosema ceranae, varroa mite infestation rate on bees = varroa infestation rate; beebread: 292 residues of pesticides). In addition, colony strength in autumn and the beekeeper's experience was recorded. Statistical analysis was performed using a General Linear Mixed Model (binomial distribution) with a stepwise forward selection. Two of the nine tested factors were included in the final model. High varroa infestation increased the probability of loss significantly ($p < 0.001$). DWV was correlated with varroa infestation rate ($r = 0.42$) and thus did not increase the fit of the model. Surprisingly, the absence of CBPV was correlated with high colony losses ($p = 0.032$). There was a trend that a high number of insecticides in bee bread was correlated with low winter losses ($p = 0.051$). Two pathogens (N. ceranae, ABPV) and colony strength showed no significant correlation with winter losses. The results confirm other European studies, that the varroa mite in combination with a DWV infection is a main risk factor for winter loss. It also demonstrates the difficulty of interpreting modelling results, when these are contradictory to the general expectation as with the effects of CBPV and of the number of insecticides. We suggest that these factors are correlated with beneficial factors (colony strength, diverse and high-quality food sources, inhibitory effects between viruses), which overrule the harmful effects of the mentioned factors.

[P.15.351] Presence, quantification and replication of DWV, BQCV and VDV-1 in the stingless bee, *Melipona colimana*, from Jalisco, Mexico

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Melipona colimana is a stingless bee endemic to the temperate region of Jalisco, Mexico, where it provides pollinating services and is cultured for the production of honey and wax. Thus far, there is no information about the sanitary status of M. colimana. However, the proximity of M. colimana to honey bees (*Apis mellifera*) that are managed in beekeeping operations, could make pathogens transmission between the two species possible. We diagnosed the presence of three of the most common honey bee viruses, deformed wing virus (DWV), black queen cell virus (BQCV) and Varroa destructor virus-1 (VDV-1), in samples of both, honey bees inhabiting in close proximity to M. colimana colonies and M. colimana samples. *A. mellifera* and M. colimana were positive to DWV and BQCV, but negative to VDV-1. Quantification analyses of the positive samples showed that *A. mellifera* had significantly higher levels of DWV and BQCV compared to M. colimana. Additionally, evidence of DWV and BQCV replication in M. colimana was found by visualizing the minus-strand of DWV and BQCV RNA using tagged primers for RT-PCR. This is the first report of viral levels and replication of DWV and BQCV in M. colimana. Due to the importance of M. colimana for regional ecosystems and its use in local beekeeping operations, the potential impact of honey bee viruses in M. colimana populations deserves further investigation.

[P.15.352] The Dynamics of Deformed Wing Virus Concentration and Host Defensive Gene Expression after Varroa Mite Parasitism in Honey Bees, *Apis mellifera*

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The synergistic interactions between the ectoparasitic mite Varroa destructor and Deformed wing virus (DWV) lead to the reduction in lifespan of the European honey bee *Apis mellifera* and often have been implicated in colony losses worldwide. However, to date, the underlying processes and mechanisms that form the multipartite interaction between the bee, mite, and virus have not been fully explained. To gain a better understanding of honey bees' defense response to Varroa mite infestation and DWV infection, the DWV titers and transcription profiles of genes originating from RNAi, immunity, wound response, and homeostatic signaling pathways were monitored

over a period of eight days. With respect to DWV, we observed low viral titers at early timepoints that coincided with high levels of Toll pathway transcription factor Dorsal, and its downstream immune effector molecules Hymenoptaecin, Apidaecin, Abaecin, and Defensin 1. However, we observed a striking increase in viral titers beginning after two days that coincided with a decrease in Dorsal levels and its corresponding immune effector molecules, and the small ubiquitin-like modifier (SUMO) ligase repressor of Dorsal, PIAS3. We observed a similar expression pattern for genes expressing transcripts for the RNA interference (Dicer/Argonaute), wound/homeostatic (Janus Kinase), and tissue growth (Map kinase/Wnt) pathways. Our results demonstrate that on a whole, honey bees are able to mount an immediate, albeit, temporally limited, immune and homeostatic response to Varroa and DWV infections, after which downregulation of these pathways leaves the bee vulnerable to expansive viral replication. The critical insights into the defense response upon Varroa and DWV challenges generated in this study may serve as a solid base for future research on the development of effective and efficient disease management strategies in honey bees.

[P.15.353] Evidence of widespread presence and replication of honey bee viruses among wild bee pollinators

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Managed and wild bees are valuable pollinators of flowering plants, but their populations have been declining in recent years, and thus, it is critical to study the potential drivers of these declines. Honey bee viruses have been associated to cases of colony losses and are candidates for interspecies transmission because they evolve fast, which could potentially threaten pollinator populations if they jump to other hosts. We investigated the presence of honey bee viruses in eight species of bee pollinators including honey bees, bumble bees, stingless bees, and solitary bees that are widely distributed in Mexico. Samples were collected from flowers in three states and the areas of collection were within 1 km range from an established honey bee apiary. The presence of the following viruses was determined using RT-PCR: deformed wing virus (DWV), black queen cell virus (BQCV), acute bee paralysis virus (ABPV), Israeli acute paralysis virus (IAPV) and Kashmir bee virus (KBV). DWV and BQCV were detected in *A. mellifera*, as well as in 50-100% of the samples of four stingless bee species, two Bombus species and one solitary bee species. None of the samples were positive for IAPV, SBV and ABPV. The sequences of DWV and BQCV were blasted against GenBank sequences and identity was >97%. Additionally, Minus DWV RNA strands were detected with tagged RT-PCR in all samples, indicating that the virus replicates in the eight pollinator species. This is also the first report of honey bee viruses found in six wild bee pollinator species. Our findings support the notion that the transmission of viruses between bees of different species is widespread and that at least one of them replicates in numerous species. If pathogenic to the bees, viral infections could result in negative impacts in agricultural and wild ecosystems.

[P.15.354] Treatment and Supplemental Feeding on Colony Production and Survival In Canada

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We examined what effect management factors, specifically treatment for Nosema spp. with fumagillin and provision of supplemental nutrition, had on honey bee colony productivity, pathogen levels, and survival. The study was conducted at three locations: 125 colonies in northern Alberta, Canada, managed for honey production; 120 colonies in southern Alberta managed for hybrid canola seed pollination; and 76 colonies on Prince Edward Island, managed for lowbush blueberry pollination. Protein patty supplements with 25% pollen were provided to half the colonies continuously during the active beekeeping season, except during the bloom period of the major crop. Fumagillin was provided in sugar syrup to half the colonies each fall. Colony populations were assessed at 11 time intervals between May 2014 and May 2016, and samples of adult bees were collected for determination of parasite, pathogen and virus loads.
Sites differed in colony survival over two years, adult and brood populations, and seasonal population patterns. Colony performance in blueberries was poorest overall. Supplemental protein feeding had no general effect on colony productivity measurements and neither protein supplementation nor fumagillin treatment influenced overall colony survival at the two-year time point. Treatment with fumagillin did suppress Nosema spp. loads in colonies after treatment, but was only associated with improved rates of winter survival and honey production for specific site-year combinations. Overall, our results provide modest, conditional support for the use of protein supplements and fumagillin.
Parasite loads in adult bee samples showed that colonies originating from New Zealand packages were initially predominantly infected with N. apis., however after one year, N. ceranae was found to be the dominant species. The trypanosomatid *Lotmaria passim* was found to be common among colonies. Increased frequency of L. passim detection over the experiment was also associated with greater infections of Nosema spp., as well as increased colony weight loss during winter. Overall, increased incidence of clinical disease symptoms in colonies were found to be highly associated with decreases in honey production and reduced fall adult bee populations, suggesting that disease status may be the most influential determinant of colony outcomes in cold temperate climates.

[P.15.355] Winter colony loss rates in New Zealand: Survey evidence and trends

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The 2018 New Zealand Colony Loss Survey estimates losses over winter 2018. It builds on 2015, 2016, and 2017 surveys, providing an opportunity for monitoring both national-level and regional-level losses over time in a country where the number of colonies has more than doubled since 2012. The survey questionnaire was adapted from the COLOSS survey to include questions of interest to New Zealand and was administered online.
In total, 3,655 beekeepers completed the 2018 survey, indicating a response rate of 47.1% of all registered beekeepers with valid email addresses. Together, these beekeepers reported on 365,986 production colonies, 42.5% of all colonies registered with an email address (and 44.0% of all colonies registered to commercial beekeepers). This poster reports on overall loss rates and perceived causes over time. The overall loss rate is estimated to be 10.21%, with a 95% confidence interval of [9.85%, 10.58%], statistically indistinguishable from those in 2016 and 2017. However, overall loss rates for winter 2018 are significantly higher than for winter 2015, providing evidence that loss rates have increased nationally.
Overall loss rates show strong regional variation, ranging between 8.06% [7.45%, 8.71%] for the Lower North Island and 12.82% [12.00%, 13.68%] for the Upper North Island. Overall loss rates within regions also exhibit a great deal of fluidity over time. For example, overall winter loss rates for 2018 are statistically higher than overall winter loss rates for 2017 in the Upper North Island and across the South Island; only in the Lower South Island did winter loss rates fall significantly between winter 2017 and winter 2018. Average loss rates are significantly higher for non-commercial beekeepers, although the survey results indicate wide variation in individual loss rates across geography and operation size.
Colony losses were most frequently attributed to queen problems (38.5%) and suspected varroa and related complications (23.1%), followed by suspected starvation (9.3%), and wasps (9.2%). Losses were also frequently attributed to suspected nosema and other diseases (4.9%) and robbing by other bees (3.5%). Competition from other beekeepers and inadequate floral resources are emerging as important concerns, particularly in manuka-rich areas.

[P.15.356] The German Bee Monitoring (DeBiMo): Report 2017/ 2018

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The German Bee Monitoring is a long-term project that focuses on the monitoring of winter losses of honey bee colonies in Germany and the impact of bee diseases, pesticide residues and beekeeping management on these losses. For the winter 2017/2018, we will present and discuss the prevalence of different pathogens , residues of pesticides in bee bread and winter mortality based on data from 1.052 bee colonies in 109 apiaries and compare the data with results from previous years.
During the winter 2017/2018, 12.6 % of the monitored colonies (N=1.052) died. These winter losses were slightly higher compared to the losses of previous years and were confirmed by reports from German beekeeping organizations. Bee samples that were taken from the monitored apiaries in autumn 2017 (N=541) revealed a prevalence of deformed wing virus of 17.7 %, whereas acute bee paralysis virus could be detected in 7.8 %, sacbrood virus in 1.1 % and chronic bee paralysis virus in 1.5 % of the samples. Our data again confirm that the infestation level with Varroa destructor and the infection with deformed wing virus in autumn are significantly correlated with the winter losses of the monitored honey bee colonies whereas the infections with Nosema spp. did not have a measurable impact on the overwintering.
We further tested 130 samples of bee bread for 451 different pesticides and detected 90 agents, most of them in traces. Only 10 samples (7.7%) were free of measureable residues (LOD 1-5 µg/kg). We found up to 33 different substances in one sample with a mean of 6 different substances per sample. The most frequent pesticides originated from applications in flowering oil seed rape. However, no differences in overwintering were observed between apiaries with high or low number of pesticides. Therefore, an impact of the here identified mixture of pesticides in bee bread collected during spring and summer and winter losses could not be affirmed so far. According to these results, Varroa destructor and the associated viruses are still the main drivers for winter losses in Germany.

[P.15.357] Another Look at Social Fever in Honey Bees

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Honey bees use a number of strategies to protect themselves and the colony from parasites and pathogens. Separate from individual immunity, social immunity is a cumulative effort of individuals to limit the spread of parasites and pathogens through the colony. Examples of social immunity in honey bees that have received attention in recent years include hygienic behavior, or the removal of diseased

brood, and the collection and deposition of antimicrobial resins (propolis) on nest surfaces. Advances in our understanding of another form of social immunity, social fever, have been lacking. Honey bees have been shown to raise the nest temperature in response to the temperature-sensitive brood pathogen, Ascosphaera apis. The modest increase in nest temperature (~0.5ºC) is thought to limit the growth and spread of A. apis to uninfected brood. We established observation hives and monitored temperature in the brood nest for approximately 30 days. The 30-day time course was separated into four distinct experimental periods: pre-exposure, sucrose feeding, A. apis exposure, and post-exposure. During the A. apis exposure period, hives were given a 1% solution of sporulating chalkbrood mummies in sucrose. Similar to previous reports, we observed a modest increase in nest temperature after A. apis exposure. However, all hives presented symptoms of chalkbrood, suggesting that elevation of nest temperature was not sufficient to limit the establishment of infection. We also began to explore the molecular mechanisms of temperature increase by exposing adult bees in cages to A. apis. Compared to bees given sucrose only, bees exposed to A. apis showed an increase in expression of an antimicrobial response peptide, abaecin, but expression of a heat stress protein was unaffected. These results indicate that honey bee adults exposed to a brood pathogen elevate the temperature of the nest and initiate an immune response, but the effect of the fever on reducing disease requires further study.

[P.15.358] The lifespan, immunity and resistance to oxidative stress of honey bees is affected by co-infection of gut parasites Lotmaria passim and Nosema ceranae

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The trypanosome *Lotmaria passim* appears to be the predominant trypanosome in honeybees worldwide. There are recently new reports of its prevalence in different country, but there are not information about its potential impact on bee survival and immunity, single or co-infected with other gut parasites. The bee survival, parasite loads, the expression of the antimicrobial peptide (abaecin, defensin and hymenoptaecin) and vitellogenin gene (linked to resistance to oxidative stress) were measured. Thus, newly emerged bees (2 days old) were maintained under controlled condition according to the follow treatments: (1) bees naturally infected with L. passim from hives infected with L. passim, (2) healthy bees inoculated with Nosema ceranae, (3) bees naturally infected with L. passim and inoculated with N. ceranae, and (4) healthy bees (control). Healthy bees inoculated only with N. ceranae and bees with L. passim and co-infected N. ceranae showed a survival under 50% and 30%, respectively. On the contrary, the survival of bees naturally infected with L. passim was not different from control. This effect could be associated to the parasitic load since that N. ceranae load was higher (3.5 x 108 to 9.0 x 108 spores per bee) than L. passim (1.3 x 102 to 1.4 x 105 cells per bee) at 20 days post-inoculation (dpi). Bees naturally infected only with L. passim did not alter the expression of abaecin and hymenoptaecin, which were similar to control at 10 dpi. On the other hand, the expressions of these genes were increased in bees infected only with N. ceranae and with L. passim and co-infected with N. ceranae in this period. Contrary, the expression of abaecin, hymenoptaecin, defensin and vitellognin were decreased drastically at 15 dpi, especially in bees infected only with L. passim or co-infected with L. passim and N. ceranae. The decreasing of antimicrobial peptides and vitellogenin expression in this period in co-infected bees was consistent with the reduced survival that was observed in this study, indicating the co-infection by L. passim and N. ceranae may cause a synergic effect at long-term, affecting the lifespan, the immunity and resistance to oxidative stress of worker bees.

[P.15.359] Effect of the concentration of toxic honeydew caused by the River Disease in honeybees on larval survival

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The River Disease is an intoxication caused by a honeydew which produces massive mortality of honeybees larvae. The intensity of this mortality defines the clinical presentation which can be serious where all the larvae die and the moderate or mild ones where a part of them survives and is operculated. The objective of the work was to establish the larval survival according to the concentration of the toxic honeydew offered to the colony. Twenty bee packages of bees were used each of them was installed in a nucleus with 3 combs, an internal feeder and a pollen cake of 500 g. The 20 colonies were divided into 5 groups. A colony of each group was housed in 4 tulle tents of 4 x 6 m and they were offered 2 Kg of toxic honeydew / normal honey with the following concentrations according to the group: I) 0%, II) 12.5 %, III) 25%, IV) 50%, and V) 100%. The toxic honeydew used came from colonies affected by River Disease and was typified by the QSI laboratory (Germany). Daily monitoring of larval development was carried out by means of photography and larval survival was measured by image analysis. The % of larval survival in each group: I) 91.3 ± 4.3, II) 63.1 ± 20.5, III) 18.4 ± 12.0, IV) 0.5 ± 0.6, and V) 0.0. The toxic honeydew produces larval mortality at low concentrations and the % of larval survival is inversely proportional to the concentration of the toxic honeydew. In field conditions the different clinical presentation of River Disease could be explained by the relative weight of the toxic honeydew on the other resources that the colony has or exploits. On the other hand it sustains the management of total harvest the colonies affected by River Disease when the agent’s admission ceases.

[P.15.360] Finding immune responsive proteins in the honey bee

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Honey bees are susceptible to a large range of parasites and pathogens, and infections can significantly affect colony health and productivity. Pesticide and antibiotic treatments are used to control disease outbreaks but are unsustainable for long term management. As pests evolve resistance towards these treatments, this results in unintentional selection of more virulent pests and less resistant bees. Consequently much research has focused on improving bee stock by selectively breeding for disease resistant traits. Once infected with a disease, bees mount innate immune responses, comprised of cellular and humoral defences. Therefore, breeding for immune competence would also increase disease resistance. This project aims to develop an immune response assay, providing a method for immunocompetence evaluation. Here we present findings from our initial immune response assay towards the fungal pathogen Nosema apis. Male honey bees were infected with N. apis and analysed using targeted protein quantitation through multiple reaction monitoring (MRM) mass spectrometry. Once further tested in other diseases, the panel of markers can be used to study immune response variations, potentially providing a platform for marker-directed breeding of more immune competent bees.

[P.15.361] Distribution and impacts of Aethina tumida Murray (Coleoptera: Nitidulidae) in Latin America

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The small hive beetle (SHB) Aethina tumida is a parasitic pest and scavenger of social bees, native to sub-Saharan Africa. Although in its native range it does not cause severe damage to strong colonies, it has invaded new areas, such as the USA and Australia, causing significant economic losses. Here, we present the current distribution of the SHB in Latin America, and discuss the strategies carried out in different countries to prevent its entrance or spread. The first report of its presence dates to 2007, in Coahuila, Mexico, a borderline state to the USA. Then it spread to other Mexican states and by 2012, it was detected in Yucatan and Quintana Roo, in the southern part of the country. Simultaneously it was also detected in Cuba, and continued spreading south, being found in El Salvador in 2013, Nicaragua in 2014 and Costa Rica in 2015. It was also detected in Sao Paulo (southeast of Brazil) in 2015, and in Río de Janeiro the following year; reaching by 2019 Mato Grosso do Sul (Midwest of Brazil). Although A. tumida does not significantly impact beekeeping in countries where Africanized bees are present, it negatively impacts the regulation of the hive products from these countries.
On the other hand, in the Andean and southern portion of South America (Bolivia, Perú, Chile, Argentina and Uruguay), there are to date no reports of the beetle`s presence. A workshop held in 2016 in the framework of RedLAC (Network for the development of familiar agriculture in LatinAmerica and the Caribbean), brought researchers and technicians of different countries to discuss strategies for the prevention and control of the beetle. The documents produced at this meeting were an important input for the animal sanitary agencies. Since then, different strategies have been drawn for its early detection, including sampling in risk areas, installation of sentinel apiaries with traps, and/or extension activities for beekeepers, among others. Within RedLAC and SOLATINA (Latin American Society for Bee Research), we aim to work together to strengthen and improve these strategies, while understanding the potential impacts of this pest for the bees in the region.

[P.15.362] Varroa mite impacts on queen bee quality in the Hawaiian archipelago

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In the midst of widespread pollinator declines, including high annual losses of *Apis mellifera* (honey bees), beekeepers struggle to pin-point the exact cause of their loss. While many factors impacting *A. mellifera* declines have been studied, little is known about the impact of the

Varroa destructor mite on the mating frequency and quality of queen bees. The Hawaiian Islands offer the unique possibility to examine the impact of Varroa on the same bee stock in the same environment by comparisons between mite-infested and mite-free islands. I mated queen bees on four Hawaiian Islands: Kauai, Oahu, Maui, and Hawaii island, and collected their offspring to determine queen mating frequency and mating quality. Using microsatellite DNA markers, worker bees were genotyped for the number of paternal lines in each of the 37 colonies tested. No significant difference for mating frequency was found between the islands with Varroa mites and islands without, and high polyandry was detected overall. This may suggest that Varroa mite presence in tropical climates is not as impactful as previously thought. We did find a significant difference in the sperm count among the islands, where sperm count correlated with managed colony numbers in areas where queens mated. This information gives us insight on Varroa mite impacts on genetic diversity and other major factors contributing to colony decline.

[P.15.363] Prevalence of Acarapis woodi in swarms of bees with high and low Africanization level captured in Mexico's high plateau

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Acarapis woodi is a mite that parasitizes the tracheae of honey bees. The mite was reported for the first time in Jalisco, Mexico in 1980. By 1983, it was found in 31 of 32 states due in part to the dispersion of swarms. One-hundred samples of swarms captured in the Valley of Mexico (high plateau) were analyzed. Half of the samples had been classified as highly Africanized and half as low Africanized based on morphometric analyses of their bees (FABIS I and FABIS II). Samples of bees from these swarms were diagnosed for tracheal mites by dissecting the tracheae from thoracic muscles and by rinsing them with 100% lactic acid, after which they were observed under a stereoscopic microscope. The Xi2 test was used in swarms with high and low levels of Africanization. Parasitized and non-parasitized swarms were evaluated with A. woodi. The following hypotheses were stated for Xi2, H0: Africanization does not influence the susceptibility to A. woodi and H1: Africanization does influence the susceptibility to A. woodi. In swarms of bees with high level of Africanization, 5 were positive and showed an intensity of 2 to 22 mites per bee. In swarms of bees with low level of Africanization, 18 were positive and showed an intensity of 1 to 169 mites per bee. The expected theoretical frequency (tf) for the data of the 5 high-level swarms of Africanization and the 18 swarms of low level of Africanization of both parasitized groups of bees was 11.5. The expected theoretical frequency (tf) for the data from the 45 swarms with high-level of Africanization and the 32 swarms with low-level of Africanization that were not infested with parasites was 38.5. The degrees of freedom of the test was 1 and the result of the Xi2 = 9. 5426.We obtained a Xi2 table of 3.8415 and a calculated Xi2 of 9.5426. Since the Xi2 calculated is greater than the Xi2 table, the null hypothesis or H0 is rejected. Hence, Africanization does influence susceptibility to A. woodi. This experiment showed that Africanization influences the level of A. woodi infestation in swarms of bees.

[P.15.364] New players in honey bee immunity: the role of reactive nitrogen and oxygen species

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Honey bee immunity is influenced by many factors, such as pests and pathogens, pesticides, environment and also beekeeping practice. Beside the social immunity displayed by honey bees at the colony level, individual bees are also equipped with a complex network of closely interconnected cellular and humoral immunity. Similarly to previous observations on other insect species, reactive nitrogen (RNS) and oxygen (ROS) species have emerged as important players involved in molecular mechanisms of bee immunity. These highly reactive compounds are known to be involved in signalling cascades following the pathogen recognition and also as effector molecules providing antimicrobial action in immune responses on local and systemic levels. Within the insect epithelial immunity, upregulated production of ROS and nitric oxide (NO) in the early phase after pathogen infection stimulates expression of antimicrobial peptides through Toll signalling pathway as a part of local responses to invading microbes. Furthermore, ROS- and NO-signalling pathways interact within the activation of systemic responses through various components of cellular and humoral immunity, including the fat tissue and hemocytes.

Actual knowledge about functions and importance of ROS and NO in honey bee immunity and health is quite limited. Our research combines biochemical, immunochemical and microscopic methods to study levels of ROS, NO and enzymes of their metabolism in bees or bee tissues challenged with relevant microbial pathogens. We have performed a detailed characterization of key enzymes of ROS metabolism, NADPH oxidase, superoxide dismutase and catalase, and of NO metabolism, nitric oxide synthase and S-nitrosoglutathione reductase, in bees infected with model bacterial pathogens. Furthermore, we initiated a pilot study on the role of ROS and RNS in bees infected with selected viruses. Finally, we aim to investigate how ROS- and NO-dependent immune mechanisms might be modulated by bee nutritional status or seasonal factors. The obtained results can contribute to further understanding of the regulatory and effector roles of ROS and RNS in mechanisms of bee tolerance or resistance to biotic stress factors.

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[P.15.366] Apiary sentinels for the early detection of Aethina Tumida in Costa Rica

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The Small Beetle of the Hive (PEC), which is native to the African continent, is a health problem for *Apis mellifera* bees, and like many other pests has spread to different countries in the world. In Central America so far, reports have been made of the presence of the PEC to the OIE in El Salvador, Nicaragua and Costa Rica.
Among the methodologies for the early detection of pests are the monitoring of active surveillance in border areas using sentinel apiaries. In the case of Costa Rica, in December 2014, a sentinel apiary consisting of three hives of *Apis mellifera scutellata* was placed, with hygienic behavior in La Cruz de Guanacaste, approximately 20 km from the site where the PEC was detected in Nicaragua. The inspection of these apiaries was carried out every month using direct observation, reviewing the whole of the hive, for this inspection the lid was first observed, then frame was taken out by frame and finally the bottom of each hive was checked. In addition, Beetle Blaster © traps (with cooking oil) were used, which facilitate the early detection of the beetle. These monitoring culminated with the detection of the PEC by SENASA officials in August 2015, with its subsequent report to the OIE. For 2016, traps are placed in the first production apiary located at 37.42 km from the initial sentinel apiary, so far its presence has not been reported.
In February 2017, four sentinel apiaries were located in the canton of Upala (northern area of the country). Two Beetle Blaster © traps with cooking oil were placed in each hive. From its placement each apiary was inspected every two months and to date the beetle has not been detected.
In December 2018, the PEC presence was detected in a sentinel apiary located at 25.76 km from the initial focus. This has allowed us to observe the progress of the plague and is currently continuing monitoring.

[P.15.367] Parasites and predators of honey bee *Apis mellifera adansonii* in Benin

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The objective of this study was to identify parasites and predators of bees in the context of bee diversity preservation. Bees were sampled from thirty (30) beehives belonging to the three climatic zones. In each study apiary, two to three hives were examined and 100 to 300 bees were collected in small bottle containing 70% ethanol. Thereafter, the hives were examined for the presence or absence of other parasites and predators. The results showed that the major pests and predators recorded were as followed: Varroa destructor (Anderson & Trueman, 2000). It has a double effect because of its parasitic predation and infectious contamination. Aethina tumida (Murray 1867) or the small hive beetle was the second by importance. The, followed, *Galleria mellonella* (Linnaeus, 1758) and Achroea grisella (Fabricius, 1794). These are Lepidoptera whose larvae attack only the bee brood, but very rapidly depreciating products of the hive. Braula coeca Nitzsch, 1818 (Insecta: Diptera: Braulidae) is a tiny, blind and wingless fly that lives in commensal on the bee. It is recognized as important vector viral diseases on bees. Tropilaelaps clareae Delfinado and Baker, 1962 (Acari: Mesostigmata: Laelapidae) are very small parasitic mites on bees in the hives and Acarapis woodi, which is a specific parasite, living in the trachea of the bee. Their damages are important and can be the source of desertion from the hives. Finally, Apocephalus sp. (Mesophora: Diptera) were also recorded as bee parasites in the hive in Benin. They are recognized as parasite of adult bees. This checklist, far from being exhaustive, showed that bees are threatened by many enemies. All visited apiaries were submitted to one or more parasites and predators at a time. Measures should be taken for the safeguarding of the bees.

[P.15.368] Incidence of diseases of *Apis mellifera* with special reference to Molecular characterization of European foul brood in Jammu and Kashmir, India

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The study was carried out during 2017-18 in different locations of the Jammu and Kashmir, India. The colony infection of European foul brood disease caused by Mellisococcus plutoniuswas recorded from a minimum of 6% (Pranu) to a maximum of 16.66% (Sartingal) in colonies of district Doda. Similarly, the colony infection of sac brood disease caused by sac brood virus was recorded from a minimum 5.71% in Attalgarh colonies of Doda region and maximum 33.3% in Arnia colonies of district Jammu. Percent brood infection of European foul brood was also recorded from a minimum of 1.47% (Parnote) to a maximum of 15.18% (Digdol) of district Ramban. Similarly, percent brood infection of sac brood disease was recorded from a minimum of 1.67% (Arnia colonies of district Jammu) to a maximum of 8.78% (Arbal colonies of district Banihal). Incidence of European foul brood was recorded maximum in September (14.59%) and minimum in March (1.81%). Percent brood infection of European foul brood in SKUAST – J, Chatha was recorded from a minimum of 1.81% (March) to a maximum of 15.37% (September). A polymerase chain reaction (PCR) molecular diagnosis for EFB was conducted on 60 larval samples collected from different areas of the four districts for PCR amplification of partial 16S rRNA gene fragments (486 bp). Results showed that of 60 samples only 6 samples (10%) were positive for M. plutonius in different apiaries. The study documented the occurrence of EFB by PCR assay in apiaries of different regions of Jammu and Kashmir for the first time.

[P.15.369] Investigation and identification of Black Queen Cell Virus (BQCV) in diseased apiaries in 3 provinces (Tehran, Alborz and Mazandaran) of Iran

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The Honeybee brood can be harmed by various viruses, such as black queen cell virus. The virus death the Queen Puppe. The BQC virus was originally found only in dead female Puppe that come from cells with black walls.
BQCV is a small single stranded RNA virus recently classified within the family Dicistroviridae, genus Triatovirus. Adult bee samples were collected from unhealthy bee colonies, which has been an unusual loss in bee population and significant honey bee mortality between July-September 2016-2018 from Tehran, Alborz and Mazandaran provinces of Iran. RT-PCR reaction was performed on extracted RNA from samples using the specific primers provided by Berenyi et al. (2006). Following the RT-PCR reaction an approximately 472 bp product was detected. Out of 28 samples from Tehran province, 18 samples were positive (64.28 %). Out of 47 samples from Alborz province, 24 samples were positive (51 %). Out of 38 samples from Mazandaran province, 13 samples were positive (34.21 %). We demonstrated the presence of BQCV RNA in 55 from 113 samples collected from 3 provinces of Iran. The investigation revealed that 48.67 % of the apiaries were infected with BQCV. Our study showed that the adult bee worker were important reservoir for BQCV which can be potentially a threat for queen larvae. The virus of the black Queen cell virus, alone rarely leads to bee damage. An infection is only possible if the bees are simultaneously infected with nosema apis, since these parasites destroy the intestinal wall in a mass increase. In the case of nosema apis and BQCV, the bees are much shorter-lived than if they become nosematose alone.

[P.15.370] Genetic Variation in Honey Bee Virus Susceptibility

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In recent times,multiple factors have played a role in driving honey bee colony losses, ranging from the spread of pathogens and increasing pesticides applications to habitat loss and climate change. Israeli acute paralysis virus (IAPV), one of the known bee viruses, transmitted by Varroa, is responsible for some of the collapses of honey bee colonies. Here, we present results from testing IAPV susceptibility in a diverse panel of honey bee colonies from the USDA Honey Bee Breeding, Genetics, and Physiology Research Lab and across multiple commercial U.S. queen breeders. We hypothesized that genetic variation among U.S. honey bee stocks causes differences in virus susceptibility and we predicted that IAPV inoculations of different genetic stocks under identical conditions lead to differences in survival. Honey bee queens were bred from parents tested against DWV, another honey bee virus, and thus categorized into DWV resistant, DWV susceptible, or DWV neutral groups. Worker offspring of these queens were tested for their IAPV susceptibility in a standard laboratory inoculation assay. The results indicated significant variation in IAPV susceptibility and a weak relation between DWV and IAPV susceptibility across generations. In a second study, different genetic lines of bees were obtained from multiple sources in the U.S. as follows: Two breeding lines from the USDA and one commercial breeder each from California, Hawaii, and Minnesota. Worker offspring from these queens were again inoculated with IAPV by topical applications and subsequent survival monitored. A total of 5,500 worker bees were analyzed and no significant differences were found among stocks, but significant variation within stocks among colonies was observed. In addition, using reverse transcription-PCR (RT-PCR), we quantified naturally occurring viruses in various tissues of queen bees from these genetic stocks and characterized expression of immune genes in a subsample of the experimental workers. Constitutive levels of two immune genes were significantly related to IAPV susceptibility,providing a potential mechanism for natural variation in virus susceptibility. Overall, our results inform beekeepers and queen breeders on important management decisions of different genetic stocks to help mitigate the ongoing honey bee health crisis.

POSTER SESSION 19 08:30-18:00
BEE HEALTH IV POSTER AREA

[P.19.441] Using a novel camera sensor to track Varroa transmission across the landscape

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Almost half of US hobbyist beekeepers do not treat their colonies for Varroa despite evidence that treatment reduces colony mortality. Beekeepers who do treat are concerned about bees from nearby crashing colonies traveling to their apiaries, bringing Varroa with them and vectoring mites across the landscape. This could have disastrous consequences for nearby beekeepers who have to expend more time and money to reduce mite levels after this transmission occurs. It is important to know if and to what extent this transmission is occurring to help inform management recommendations and extension activities aimed at non-treating beekeepers.
To test whether bees from crashing colonies move to neighboring apiaries, the adult bees in two nucleus colonies were painted at the University of Maryland Beltsville Research Farm in summer 2017. A colony with an extremely high Varroa load (>15 mites/100 bees) was painted red and a control colony (<3 mites/100 bees) was painted blue. When the high Varroa colony crashed, all colonies within a 3-mile radius (n=100 colonies) were searched for painted bees by visual inspection. The time required for this method became impractical, and a need to remotely monitor these 100 colony entrances was apparent. In summer 2018, a camera sensor was developed using Raspberry Pi 3B+ and Pi Camera sensors. The cameras use OpenCV to detect user-specified colors (red and blue for this experiment) and take a photo when the color is detected. Thus photos are taken of painted bees arriving at the entrances of any colony within the flight radius of the painted colonies.
A trial of ten camera units verified the cameras effectively capture painted bees. Cameras within the painted apiary captured 38 red bees from the high Varroa colony and only two blue bees from the control colony, indicating that high Varroa pressure does affect bee drift. A full trial conducted in summer 2019 will discern whether these effects translate to movement of bees between apiaries as well. We expect to find significantly more bees from the high Varroa colony in surrounding apiaries than bees from the control colony, indicating crashing colonies can transmit Varroa between apiaries.

[P.19.442] Effect of natural immune inducers on honey bee survival, Nosema ceranae spore numbers and innate immunity of Western honey bees (*Apis mellifera*)

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Nosema ceranae is the dominant microsporidium that causes nosema disease in Western honey bees (*Apis mellifera*). For several decades, the common treatment for the disease has been the antibiotic fumagillin. However, there are risks associated with this treatment, including the development of antibiotic resistance in N. ceranae and antibiotic contamination of hive products. Therefore, there is a need to develop alternative safer treatments. Four natural pathogen-associated molecular patterns (PAMPs) were examined that are known immune inducers in other insects as alternative treatments. The tested immune inducers were chitosan, peptidoglycan, flagellin and zymosan. Bees were individually inoculated with N. ceranae along with an immune inducer in sugar syrup, while the control bees were either inoculated with the pathogen or just received sugar syrup. Chitosan and peptidoglycan significantly increased honey bee survival and reduced spore numbers of N. ceranae at 20 days post treatment. Additionally, these two immune inducers influenced the expression of the genes for the antimicrobial peptides, hymenoptaecin and defensin2, as well as the gene for the stress tolerance-related protein, blue cheese, compared to the controls. These results suggest that chitosan and peptidoglycan are inducers of innate immunity and have the potential as alternative treatments for N. ceranae infections.

[P.19.443] The cardiac activity of Varroa destructor and its potential as a new bioassay for acaricide evaluation

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At the present study, we examined and described for first time the function of the heart at the acari Varroa destructor and analyzed its activity. Through light microscopy, we located the mite's heart under the dorsal exoskeleton shield and over the syngaglion, and analyzed the duration and the frequency of its activity. The heart is approximately 0.165 long and 0.06 mm wide and consists of two lateral trunks. The heart beats with an average duration of 10 sec at an average frequency of 0.13Hz under normal conditions. Monitoring the cardiac activity, we examined the impact of applied acaricides and evaluated their efficacy by recording any malfunctions following the application of treatments. We were able to evaluate with great precision the lethal dose required and the time required for each substance to act. As a result, the use of Varroa's cardiac activity has potential to be used as a new bioassay for accurate assessment of compounds presenting varroacide action.

[P.19.444] Seasonal brood interruption as an effective measure for varroa control

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Most Varroa induced colony losses occur during winter season in consequence of an insufficient health status of the winter bee population. Even with low mite infestation in early spring critical infection levels can be reached until the period of winter bee production if colonies continuously rear brood throughout the whole season. Under natural conditions, the infestation may be strongly reduced during the process of swarming. Learning from this we recommend beekeepers to artificially interrupt brood production of non-swarming colonies and developed suitable management procedures for different type of beekeeping operations. Long-term experience of German beekeepers show that the risk of colony losses and the use of chemical treatments can be widely reduced by a consequent application of such biotechnical measures.
To assess the efficacy, the workload and the impact on colony development, different methods of brood interruption were tested during 2 seasons (2016/17 and 2017/18) by 12 partners in 11 European countries. In total 472 colonies from different European honey bee subspecies were used for this study. A standardized protocol was developed and published on the COLOSS website (www.coloss.org). Queen caging (QC) for 25 days followed by a treatment with trickling of oxalic acid solution (OA - Apibioxal 4.2%) served as a standard in all apiaries and was compared with different methods' variants (2 to 5 methods/apiary), in particular total brood removal (TBR), trapping comb technique (TC), queen caging and sublimation of OA as well as other locally used control treatments.
The realized mite reduction of QC ranged from 48.6 to 95.5%. The highest efficacy was achieved with sublimation of OA in the broodless stage (average of 83.0 and 95.5%, for both seasons respectively) and with trickling of 4.2% of OA (84.0 and 86.7%). The average efficacy of the pure biotechnical, chemical free methods was estimated with 51.1% for TBR and 77.1% for TC. No major incidences related to colony development and queen losses have been observed. We conclude that a proper application of one of those brood interruption methods can strongly contribute to an efficient Varroa control and production of honey bee products with high quality and safety characteristics.

[P.19.445] A blend of Bacilli and Lactobacilli lowers Nosema spore counts in the field

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The honey bee beneficial microbiota, in addition to known pathogens, represents a key variable in honey bee colony health. Soon after eclosion, adult honeybees are inoculated with core gut microbiome species, including those belonging to genera Bacillus and Lactobacillus . Bacilli and Lactobacilli inhibit bacterial honeybee pathogens and their presence is linked with good colony health, yet majority are susceptible to antimicrobial substances used in beekeeping. Lactobacilli feeding stimulates honeybee immune response, and several commercial feeds contain Lactobacilli . Exogenous Bacilli prolong honeybee lifespan and survival of Nosema challenge, yet does not lower Nosema spore counts. We demonstrate that feeding Bacilli and Lactobacilli together significantly improves colony health and overwinter survival and is associated with lower Nosema spore counts in the field. Direct and indirect modes of action are proposed to account for the additive effects of Bacilli and Lactobacilli supplementation on Nosema infection.

[P.19.446] Aluen CAP: treat Varroa destructor with the new organic acaricide in full honeydew

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Resistance to synthetics treatment of varroa mites has increase all over the world and this has pushed to develop a new way of treating mites using organic compounds. Aluen CAP (oxalic acid in slow release strips) is a new way of treating hives against varroa, which is organic, it does not generate resistance and leaves no residue in honey. As a consequence, it allows to treat hives in full honeydew and maintain low loads of mites all over year. This leads to analyze the doses for hives with low varroa loads to reduce the active principle applied in the colonies and lower the cost of application. Efficacy of the new formulation with oxalic acid (Aluen CAP) made by Cooperativa de Trabajo Apícola Pampero Ltda. was evaluate at 4 diferent dosis: 1, 2, 3 and 4 strips per hive. The treatments were placed in big colonies (19.4 frames covered with adult bees and 8.7 brood frames on average) at the full honeydew, to test the acaricide in hard conditions. Falling mites were counted after 7, 14, 21, 28, 35, 42, 49 and 56 days using hive bottoms specially adapted for the collection of dead mites. After the last count, the strips were removed and at the same day, colonies received a chemical shock using Amitraz. The efficacy of the treatment was calculated as a percentage: ((number of dead mites during oxalic acid treatment)/(number of dead mites collected during the treatment with OA and synthetic treatment))×100. The average efficacy obtained through the trials was 33,15% (1), 76,32% (2), 83,58% (3) and 90,42% (4) at 42 days after the hives were treated, and 40,32% (1), 90,36% (2), 89,84% (3) and 95,50% (4) at 66

days after the hives were treated. These results suggest that hives with a low infestation of varroa mites could be treated with low doses of Aluen CAP, reaching a higher and slower efficacy, but lowering the cost significantly.

[P.19.447] Controlled time-release of formic and oxalic acids for control of Varroa mites

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Since its introduction to the US in the 1980s, the Varroa mite has been the most serious pest in the U.S. beekeeping industry. Over the last decade, U.S. beekeepers have lost a third of their colonies annually. Resistance has developed to earlier synthetic miticides, and these also have other significant limitations. A safe and effective natural treatment for Varroa is desperately needed.
Formic acid and oxalic acid are naturally derived miticides; resistance to these has not yet been observed. Unfortunately, current formic and oxalic hive treatments are often "homemade," resulting in variable doses, and can be hazardous to the applicator. Commercially available MAQS releases formic acid over several days, depending on hive conditions, but MAQS cannot be used in much of the country during honey production because outside temperatures are too high.
MSU has developed a polymer for controlled-release of formic and oxalic acids that has the potential to be an effective miticide in honey bee hives. The strategy involves hyperbranched poly(esters) (HBPE) from "natural", biodegradable building blocks to which the acids are covalently bonded. As the HBPE degrades, it releases the miticides in a slow, controlled rate. The release rates of these formulations have been studied in numerous laboratory trials. The rate is dependent on temperature and humidity, but the release of formic acid from the HBPE is slower than from MAQS under similar conditions, and lasts long enough to kill mites in the brood cycle, not just phoretic mites. The cost of the raw materials is low and the synthesis is simple and inexpensive.
Initial hive tests were successful, and synthesis of the miticide has been scaled up to allow testing in 54 hives. Five different dose levels and two application methods were tested, plus controls. Dosages consistent with early work remained effective in this larger study, whereas lower doses lost effectiveness.

[P.19.448] SHB management opportunities in Italy

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The first incursion of Aethina tumida Murray, the small hive beetle (SHB), in the European Union was reported in Portugal in 2004 following an importation of queen bees from Texas (USA). The stamping out measures immediately adopted prevented any further infestation and spreading of this exotic beetle. Ten years later adults and larvae of SHB were reported in Italy in September 2014 in honey bee colonies near the Gioia Tauro port in the Calabria region (southern Italy). In November 2014, an infested apiary was reported in eastern Sicily. Genetic analyses revealed the African origin of SHB introduced into Italy. Early reaction measures aimed at eradication and containment of the infestation. They required immediate notification of SHB detection to the local veterinary services, movement restriction of the concerned colonies and apiaries, destruction of the entire infested apiaries followed by ploughing and pyrethroids soil drench application. In Calabria region, 132 positive sites and a single one in Sicily were officially reported and destroyed between 2014 and 2018. The Ministry of Health granted compensation to beekeepers according to the law in force. Furthermore, a 20 km radius protection zone and a surveillance zone covering the entire territory of the Calabria and Sicily regions were established. Compulsory visits to all apiaries in the protection zone with georeferentiation and visual colony inspection according to 5% expected prevalence (95% CI) were carried out. In the surveillance zone, apiaries to be inspected were selected according to risk analysis or randomly and colonies were inspected according to 2% expected prevalence (95% CI). Sentinel honey bee nucleus colonies managed by official veterinarians were installed in the protection zone to facilitate SHB detection. In spring 2017 restriction measures applied to Sicily region were lifted while the surveillance program was continued and intensified. Additionally, the national SHB surveillance program established in spring and autumn 2015 and extended until 2018 did not reveal any SHB outside the two concerned regions. The persisting circulation of SHB in Calabria region requires a reconsideration and possibly an update of the measures adopted with a view to the future of infestation containment and beekeeping industry.

[P.19.449] Odorant receptor genes in small hive beetles

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The ability of insects to respond to chemicals in the environment is primarily determined by odorant receptor genes (Ors). Comparative analyses of the Or gene family may therefore shed light on the evolution and the relative importance of olfaction across different life histories. The small hive beetle, Aethina tumida, is a parasite of social bee colonies, and depends on olfactory cues to locate host colonies. However, it is unclear how the Or gene family evolved as an adaptation towards infesting bee colonies. Here, we investigated Or orthologues and conducted the analyses of evolutionary relationship of the Or gene family in A. tumida and three other species (*Apis* *mellifera*, Tribolium castaneum, Nasonia vitripennis) with different ecological niches. Our results show fewer Or gene family members in A. tumida. Structural analyses of odorant co-receptor (Orco) genes further suggest that A. tumida Orco is highly conserved compared with T. castaneum. These results may reflect the adaptation of A. tumida to a specialized parasitic life style in colonies and may also foster parasite control via Orco gene silencing.

[P.19.450] Effect of chitosan on the development of Nosema infection in *Apis mellifera L.* colony

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Nosema infection is one of the main causes of winter weakening and death of honey bee colonies in a temperate climate zone. Nozema has only residual mitochondrial organelles and absorbs ATP from the host's cellular environment, causing energy and oxidative stress, degeneration of the intestine and prevention of epithelial tissue renewal in infected bees. Honey bees have been shown to capable to counteracting Nosema infection by increasing nutrition. However, compensation of increased energy costs in the conditions of the long winter period and limited food resources is extremely difficult. In this case, the immune system goes to the first roles in the fight against Nosema infection. We have previously shown that chitosan of a certain molecular weight and deacetylation degree activates the protective systems of honey bee. It is noteworthy that this substance is safe and environmentally friendly, as it has a natural origin, is completely processed by the bee and does not accumulate in the beekeeping products.
We have investigated the effect of chitosan (200 kDa, 75% deacetylation) on the development of Nosema infection in bee colonies during the wintering. Autumn feeding of bee colonies with chitosan significantly reduced the infection degree of worker bees with N. apis spores and retained the barrier functions of the digestive tract until the onset of the flying period in spring. The action of chitosan also caused a magnification of humoral protection during the entire winter period. It manifested itself in an increase in the level of phenoloxidase and antioxidant enzymes activity, as well as in the expression of immune genes (vitellogenin, defensin and abaecin). In general, the use of chitosan resulted in resistance increase of bee colonies to Nosema infection and a reduction in their death during the wintering.

[P.19.451] Permanently Averting Disaster or Delaying the Inevitable? Development of a Varroa Action Plan for mite-free Newfoundland and Labrador

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Newfoundland and Labrador, Canada, is one of the few regions on Earth with a managed honey bee population that is completely free from the parasitic mite Varroa destructor. Despite provincial government importation restrictions, little in the way of Varroa-specific biosecurity measures had been developed to prevent and/or cope with a mite incursion into this last bastion of mite-free bees in North America. We report on both the process undertaken, and the progress achieved, to produce a Varroa Action Plan (VAP) for the province. The VAP is the result of collaboration by hobby and commercial beekeepers, academic researchers, government representatives, and the provincial beekeeping association. The VAP contains education efforts to prepare beekeepers for the possible arrival of Varroa destructor, biosecurity guidelines to ensure that apiaries and beekeeping activities minimize the risk of rapid mite spread across the province, research efforts towards developing mite-resistance characteristics in the bees before the arrival of the mite, and a containment and eradication plan uniquely suited to the province that may allow recovery of its mite-free status even in the event of a biosecurity breach and mite incursion. We will summarize the contents of the VAP, as well as report on findings concerning the hygienic behavior (freeze-killed assay) of a number of mite-naïve bee colonies from the province.

[P.19.452] Prevalence of honeybee viruses in hornet from China and France

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Transmission patterns and the study of virus vectors such as Varroa destructor, had drawn great concern to manage honey-bee health issues. However, little is known about the occurrence and prevalence of honeybee virus in bee predators like wasps and hornets. In the present study, we investigated the occurrence of 13 honeybee virus species in five wasp species from 4 provinces of China and 2 hornet species from 4 regions of France. The results showed that all wasp species from 9 regions of China were infected by different types of honeybee viruses and particularly most of them carried *Apis mellifera* filamentous virus (AmFV), Deformed wing virus (DWV) and Israeli acute paralysis virus (IAPV), even some were infected by more than 4 viruses simultaneously. Phylogenetic analysis on BQCV and IAPV indicated that most of IAPV strains belonged to a single phylogenetic group, while BQCV strains from China and France belonged to several groups. KV from hornet was totally different from those of *Apis mellifera*. These results could serve as a basis for further investigations of transmission and origin of honeybee pathogens in Vespidae species, especially their potential role as carrying viral vectors for bees.

[P.19.453] Glycerol may replace sucrose in oxalic acid solutions used to control Varroa infestations

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Varroa destructor is a devastating parasite of *Apis mellifera* colonies. Infestations may be controlled by effective and environmentally sound methods, such as oxalic acid (OA) solutions trickled onto the combs of broodless colonies. However, the internal ambient conditions of a hive may promote evaporation of water from the OA solution, ultimately reducing the treatment efficacy. Excipients are then required to hinder the solution dehydration.
Sucrose is typically added as a hygroscopic excipient of OA. However, the acidic environment of the solution may promote quick sucrose degradation, with unpredictable effects on treatment efficacy and tolerability. Therefore, sucrose must be considered a suboptimal excipient in OA solutions not intended for prompt use.
Pharmaceutic and food industry make wide use of glycerol as a humectant compound. Being more stable than sugars in low pH solutions, it is a promising candidate as an alternate OA excipient.
A laboratory bioassay was conducted to evaluate the concentration-effect relationship on varroa mortality of OA solutions added or unadded with 10% glycerol. Results support the use of glycerol to promote the acaricidal effect of OA.
Field trials conducted in broodless colonies during both winter and active season to compare glycerol and sucrose as excipients of OA solutions administered by trickling resulted in no significant differences on efficacy and colony conditions.
If confirmed by further trials, these results support the replacement of sucrose with glycerol as an excipient of OA products intended for the control of varroa infestations, and promote the formulation of ready-to-use OA solutions.

[P.19.454] Investigating a novel honey bee brood disease and Characterising the treatment susceptibility of US and UK strains of Melissococcus plutonius

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Colonies of honey bees in Florida have been suffering with an unknown brood disease (locally called Crud brood). This novel disease kills honey bee larvae causing a poor brood pattern and ultimately colony loss. The aim of this work was to investigate any bacterial drivers, as well as confirming the presence or absence of known pathogens, and to establish if strains isolated from the USA were resistant to the antibiotics used for brood disease therapy: OTC, Tylosin and Lincomycin. Three UK isolates with different sequence types (STs) were used as a baseline for antibiotic susceptibility. In addition, the bee bread and wax was checked for residues.
The investigations demonstrated that the disease was European foulbrood (EFB). Two different stains were detected H2A and D3A. Moreover, all samples were positive to black queen cell virus (BQCV).
Regarding the results of bee bread and bee wax analysis, a mixture of different components was found at a very high level.
Three products were provided plant extract, VITA Oxygen, natural compound, for efficacy testing against M. plutonius.
Five strains were tested (three from the UK (STs) + two USA strains). The isolate H2A was more resistant to OTC and Tylosin. D3A did not grow on any of the plates with OTC. All strains tested were susceptible to Lincomycin at the concentrations tested with the exception of ST1, the type strain.
In conclusion, the disease found was European foulbrood (EFB), in addition to black queen cell virus (BQCV).
Both H2A and D3A showed different susceptibility profiles to the UK strains of M. plutonius. H2A shows resistance to OTC and Tylosin.
Bee bread and wax samples were all positive for at least 2 compounds. Some pesticides were found in all samples from hives with or without disease present.
Both Vita Oxygen at 0.3% and the natural compound at a concentration of 250ug/ml showed control of all the strains in in-vitro condition. Field trial experiments should be performed to determine the correct dosage needed in apiary settings for the potential use of both products as a control agent against European foulbrood, including the strains resistant to antibiotics.

[P.19.455] Seasonal Varroa destructor population growth in north central Florida

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A successful Integrated Pest Management (IPM) approach to Varroa destructor control must include the application of effective treatments at the correct time. Correspondingly, we conducted an experiment using four groups of honey colonies maintained in different apiaries in north central Florida to understand the effect that the timing of treatment application has on Varroa populations at different seasons. Each apiary was assigned to a specific season and consisted of 16 – 20 honey bee colonies. The apiary sites were as follows: 1) winter – 20 hives located in Gainesville, FL, USA, 2) spring – 20 hives located in Citra, FL, USA, 3) summer – 20 hives located in Hawthorne, FL, USA, and 4) fall – 16 hives located in Gainesville, FL, USA. Prior to the start of the experiment, colonies within each apiary were treated with acaricides (amitraz + oxalic acid) during their assigned season (winter, spring, summer, fall) to bring Varroa populations as close to zero (< 0.4 mites/100 bees) as possible. Following this, mite populations (# mites/100 bees) were monitored monthly via alcohol washes. Monitoring of Varroa continued for each group until the mite population peaked and subsequently declined for two consecutive months. Using #mites/100 bees, we calculated the Varroa population doubling rate and compared the slopes of the population increases across the four groups. Our results will serve as a foundation for Varroa treatment models, thus aiding beekeepers in the future as they develop an IPM approach to control Varroa.

[P.19.456] Evaluation of 3 methods of oxalic acid application, liquid, heat vaporization and spray, in controlling varroa mites

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The use of oxalic acid for controlling varroa mites in proven necessary, as these mites are becoming more resistant to treatments of pyrethroids. The purpose of this research was to find an effective, efficient and economic solution, as these mites carry many viruses, which can spread quickly, causing the entire bee colony to collapse. In the period of 3 years, 2016-2018, 3 methods of oxalic acid application – liquid, heat vaporization and spray – were used during winter, when the colony was broodless, as well as in spring, side effects, efficiency, and the success rate of each method were compared. 400 hives were divided into 4 groups of 100 hives each, the average percentage of varroa mites were calculated by alcohol wash procedure, as %6 in September 1st. During winter, in November and December: In group 1, heat vaporization application was used 25 times, each for 1 minute. In Feb 10th, the level of varroa mites was at 0.10% and by August 23rd, it reached to 0.60%. No adverse side effects were found on the bees. In group 2, liquid form was used, feeding the bees a mixture of oxalic acid and sugar syrup. After 2 times, bees' population began to reduce. So, this method should be used only as the last alternative. On February 10th, the level of varroa mites was 1% and on August 23rd it reached to 12%. In group 3, the spray method was used 4 times. On February 10th, the level of varroa mites was at 0.20% and by August 23rd, it reached to 3%. In group 4 where no treatments were used, the level of varroa mites in February 10th, was 6% and in May 25th it was 24%. 80 of the infested hives removed to be treated and from the 20 hives that remained 16 died before August 23rd. The remainder 4 hives were highly infested, over 60%. Heat vaporization and spray proven to be the effective methods.

[P.19.457] On the natural cell size of European honey bees: fatal error or distortion of historical data?

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As a possible way to help control varroa mites, some beekeepers advocate the use of cells smaller than the regular size commonly used by beekeepers. This paper addresses two of their principal arguments, namely that honey bees built smaller cells under natural conditions in the past, and that a fatal error occurred at the turn of the 20th century when a new and allegedly misleading method of estimating cell density was introduced. Historical data show not only that cell sizes were not smaller in the past, but also that estimating cell densities was not an issue before the introduction of wax foundation. Moreover, not realizing that the two methods of estimating cell densities are equivalent, the proponents of small cells have erroneously corrected the data reported by the authors of the 17th, 18th and 19th centuries. In conclusion, the claim that cells were smaller in the past is not only not supported by the historical records, but rests on a distortion of the historical records resulting from an incorrect transformation of the original data.

[P.19.458] An optimised method of varroa treatment with formic acid: a comparative approach 2018-2019

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Varroa destructor (Acari: Varroidae) is the ectoparasitic mite of the honey bee which causes important damages in beekeeping. Its reproduction characteristics connected to capped brood lead to a general poor efficiency of actual treatments which are limited to kill mites on adult bees. The need to cut the life cycle is ever increasing, also taking into account the new findings which emphasize the destructive capacity of varroa that feeds primarily on fat body (Ramsey et al., 2019), which affects the health status of honeybees in all developing stages. Actually, there are different methods used for the varroa treatment with formic acid with various degree of efficiency, most of them being completed by classical treatments for a better control. The concentration, quantity and application methods as well as their place in the colony are quite diverse. To shorten the term of fumigation with formic acid on honeybee colonies in order to limit the side effects on bee colony with a good efficiency in killing varroa inside the capped brood in the same time with those found on adult bees is an important goal of actual researches (van Engelsdorp et al., 2008, Siceanu et al., 2018, Siceanu et al., 2019.The objective of the study was to establish the effect of formic acid on varroa, inside the capped brood cells by different treatment methods. The experiments were carried out in 2018-2019 on honeybee colonies highly infested with varroa, in a research apiary in Romania. The results show that the most protonympha and deutonympha in the cells are killed by formic acid after comparative experiments 2018/2019, which show that these treatments cut the life cycle of varroa in its reproduction phase. The other stages (male, mother foundress, daughter) can suffer, but the variation of mortalities is relatively large (from 0 to 100%), the variation being caused probably by how well they are exposed to formic acid and the degree of exoskeleton chitinization, these stages being less susceptible to treatment.

[P.19.459] BPRACTICES project: New indicators and on-farm practices to improve honeybee health in the Aethina tumida era in Europe

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BPRACTICES is a project funded by the European Union's Horizon 2020 research and innovation program under Grant Agreement n° 696231, ERA-Net SusAn – European Research Area on Sustainable Animal Production Systems, that aims to develop a sustainable beekeeping system by implementing innovative management practices (Good Beekeeping Practices - GBPs). The project consortium, coordinated by the Istituto Zooprofilattico Sperimentale del Lazio e della Toscana "M. Aleandri" (Italy), includes: University of Namik Kemal (Turkey), Agricultural Institute of Slovenia (Slovenia), Centro de Investigación Apícola y Agroambiental de Marchamalo (Spain), Austrian Agency for Health and Food Safety (Austria), and Istituto Zooprofilattico Sperimentale delle Venezie (Italy). Moreover, the project involves: the International Federation of Beekeepers Associations (APIMONDIA), the University of Genova (Italy), the Danish Beekeepers' Association, the Appalachian State University and has the valuable collaboration of the European Union Reference Laboratory for Bee Health (ANSES, France) and of the Food and Agriculture Organization of the United Nations (FAO) Technologies and practices for small agricultural producers (TECA) platform. Within the project, a list of general and specific GBPs to prevent and control the main honey bee diseases has been set up. Sustainable organic compounds and biotechnical methods for diseases control have been identified in order to promote a sustainable and resilient beekeeping sector. Their application in the beekeeping sector to enhance honey bee health, will reduce the use of chemicals, increasing quality and quantity of bee products. Innovative laboratory diagnostic methods have been tested (ring test) in order to provide efficient preclicinical diagnostic methods for bee diseases (e.g. PCR analyses from innovative matrices) validated and standardized at international level. Biosensors to detect contamination by pesticides and the presence of pathogens are being developed. An innovative informative technology (QRCode/RFID system) has been developed to enhance consumers' trust and acceptability. All research outcomes have been validated under practical conditions. An economic evaluation of competitiveness and resilience of European beekeeping is ongoing thanks to the valuable collaboration of APIMONDIA. Consumers' data will be collected to evaluate the system and communication activities are ensuring the visibility of the project for all the beekeeping sector stakeholders.

[P.19.460] Developmental thresholds of immature stages of Small Hive Beetle (SHB) in South Korea

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Small Hive beetle (SHB), Aethina tumida Murray, (Coleoptera: Nitidulidae) is an invasive pest of honeybees colonies in the South Korea. We investigated influence of temperature on immature stages of SHB. The Immature stages were exposed to different temperatures (15, 20, 25, 25, 30, 35oC). The shortest development of eggs, larvae and pupae were on 35oC. While, developmental period of immature stages was highest at 20oC. The development was not completed at 15oC. Maximum and minimum survival of immature stages was recorded at 25 and 35oC, respectively. The developmental thresholds were calculated by linear model. The development threshold of egg, larvae and pupae were 9.0, 12.85 and 9.92 days, respectively. The thermal constants for development were 31.54, 96.51 and 357.14 degree-days for eggs, larvae and pupae, respectively. These developmental thresholds can be used to predict potential distribution of Small hive beetle in South Korea.

[P.19.461] A Varroa Mite IPM Program for New England Honey Beekeepers

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In New England (USA), an estimated 35-45% of crops rely on insect pollination for seed and fruit set making honey bees vital for agriculture in the region. Unfortunately, like most areas, beekeepers continue to experience significant colony losses. During the 2016/2017 season, beekeepers reported losses around 53% in both Massachusetts and Maine, far surpassing national averages. Of all the problems facing

honeybees, the Varroa mite (Varroa destructor) continues to be the most common cause of hive loss.
An Integrated Pest Management (IPM) based approach (including monitoring, using thresholds to make decisions, prevention, intervention, etc.) provides the best control of Varroa populations within hives. While a variety of options exist for monitoring and controlling Varroa mites, there is a serious deficit in the availability of information, education, and training on creating and implementing an IPM program. Furthermore, New England has unique management challenges (shorter spring/summer colony buildup season and harsh winter conditions) that other areas of the Unites States do not face, so many of the of the current education/extension/management materials are not applicable to beekeepers in New England.
The foundation of IPM is monitoring pest populations. In the 2016/2017 beekeeper survey between 25-32% beekeepers monitored mite populations to determine if treatment thresholds have been reached or efficacy of the applied treatments. The majority of beekeepers continue to use a calendar to determine when to apply treatments.
The Varroa Mite IPM Program for New England Beekeepers project was created with the purpose to promote the development and adoption of IPM based protocols for Varroa mite by providing beekeepers with New England focused workshops, demonstrations and protocols. Outreach efforts include the assembly and distribution of 3,000 alcohol wash jars for monitoring in Maine and Massachusetts, setting up an interactive on-line geographical based website for beekeepers to input monitoring data, and dozens of hands-on educational outreach presentations to beekeeper clubs, groups and other organizations. We will be reporting on the effectiveness of this program and presenting ideas on the way this model of outreach education can be expanded or used in other areas.

[P.19.462] LIFE STOPVESPA Project: control of Vespa velutina in Italy

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LIFE STOPVESPA (LIFE14 NAT/IT/001128 STOPVESPA - Spatial containment of Vespa velutina in Italy and establishment of an Early Warning and Rapid Response System) is an European project financed by the LIFE programme of the European Commission from August 2015 to July 2019. The project has grouped Universities, Associations, Beekeepers and Citizens with the goal to contain the spread of Vespa velutina in Italy, an invasive alien species that causes serious damage to beekeeping and biodiversity.
The actions of the project allowed to collect and process information on the presence of the Asian yellow-legged hornet and its impacts on honey bees, to evaluate pathways and drivers of introduction, to develop an early warning and rapid response system, to define guidelines for the destruction of colonial nests, to develop new monitoring techniques, to destroy nearly 2,000 nests, to produce educational material. The project has worked closely with local and national institutions and with beekeeper associations ensuring the activation of a monitoring network and emergency plans. The Department of Electronics and Telecommunications of the Polytechnic University of Turin has developed a harmonic radar to track the hornets flying back to their nests so as to detect them. The radar has been used in 2018 for the control of V. velutina diffusion in Italy, but it could find use also in several other fields of entomological research and pest management.
Besides to control activities, the LIFE STOPVESPA project has studied in these years the impact of V. velutina not only on honey bee colonies but also on native wasps, pollinators and wild bees, to fill the lack of quantitative data on the impact of V. velutina on biodiversity and pollination services.

[P.19.463] Screening of Chemical Control agent and its Field Test in Korea against a Newly Invaded Pest of European Honeybee (*Apis mellifera*), Aethina tumida Murray, 1867(Coleoptera: Nitidulidae)

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Small hive beetle (SHB, Aethina tumida Murray, Coleoptera: Nitidulidae) is a honeybee pest infesting combs and stores inside the hive. Contamination of the SHB on *Apis mellifera* colonies were firstly noticed on September 23, 2016, in Miryang City, Gyeongnam Province in Korea. We tested the preference rate of bait against small hive beetle larvae and adult. When treated each bait of preference showed highest pollen or pollen added sugar water. We was selected the coumaphos through insecticide screening. LC/LD50 values of coumaphos on small hive beetle larvae was estimated as 612ppm, and 1.591ug/beetle and fluvalinate 4,641ppm and 12.067ug/beetle. LC50 values of coumaphos on small hive beetle adult was estimated as 6,571ppm. we suggested pollen patty mixed with coumaphos 10,000ppm as a control agent against small hive beetle. The control agent was tested with CD type trap and showed 82% control efficiency after 15 days in field. This study should be used a fundamental information for developing IPM (Integrated Pest Management) and controlling the expansion of population of small hive beetle in Korea.

[P.19.464] Search for effective probiotic strains against causative agents of principal diseases of bees and study of their impact on the intestinal microbioce-nosis

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Over the last years throughout the entire world there have been a consid-erable number of deaths of honey bee colonies, which causes

severe harm to apiculture. Considering the fact that the entomophilous crop yield depends on the honey bee population, this problem goes far beyond the limits of apicul-ture.
According to the scientists, the reason for this phenomenon is a number of biotic and abiotic factors of the environment which include climatic changes, the extensive use of pesticides and cultivation of genetically modified agricul-tural crops. However, the most probable cause of the deaths of honey bee col-onies is seen in the widespread occurrence of viral, bacterial and fungal infec-tions of bees, which is related to the mass infection of apiaries with Varroa mites. As a rule, this problem is solved with the help of antibiotic medications and chemicals, which, in its turn, leads to a reduction in the quality of honey, e.g.: accumulation of antibiotics.
To solve this problem, the scientific laboratory of Bashinkom Company and the Bashkir State Agrarian University carried out a search for highly effec-tive strains in order to create microbiological medication against Penibacillus larvae and the fungi of the genus Ascosfera. The strains 11B,12B and 1K of Bacillus subtilis were isolated, they have a high antagonistic activity against Penibacillus larvae and the fungi of the genus Ascosfera. Based on the resulting strains of Bacillus subtilis, as well as of the bacteria Lactobacillus and Entero-coccus, the multistrain probiotic agents “Apivrach” and “PcheloNormoSil” were produced. The influence of the products on the intestinal microflora of bees was studied.
The use of supplementary feeding with added “Apivrach” and “Pche-loNormoSil” contributed to an increase in the amount of the sealed brood by 8.0 %, the honey yield – by 7.0 % and the wax yield – by 8.9 %, as well as an increase in the level of profitability by 6.9 %. Moreover, the group of bees that received supplementary feeding with the added agents showed a general in-crease in the disease resistance, the life span of the bees increased, the amount of the pathogenic intestinal microflora decreased considerably.

[P.19.465] Challenges with organic beekeeping; Based on researches performed in California, Washington State, Vancouver, and Iran (North, West and South)

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The focus of this presentation is to portray the challenges involved in organic beekeeping and to discuss the natural alternatives for controlling Varroa mites and treatments of diseases.
This research was performed in 6 locations in 3 countries, California, Washington State, Vancouver, and Iran (North, West and South), to gather as much information as possible for an accurate conclusion. Providing a perfect environment for this study was challenging, as they were many factors to consider such as having a low level of Varroa mites and diseased free hives (AFB, EFB and Nosema Apis) - considering that most diseases are epidemic and endemic - ensuring hives were free from chemicals and artificial supplements residues and that bees were not fed adulterated sugar; the sources of water were free from chemicals, bacteria like E.coli, and pollutants such as heavy metals. So, it became necessary to place the hives in the locations, high elevations and forests, that were distant from monoculture farms and farms that were using pesticides and chemicals; also in a proper distant from other apiaries, to prevent transfer of diseases and Varroa mites infestation.
In this research natural alternatives - oxalic acid, formic acid, and thymol- were used to control Varroa mites. The hygienic behaviors of the bees for controlling Varroa mites were closely examined; it is not recommended to rely on the hygienic behaviors of the bees as the only solution for controlling Varroa mites. To eliminate protozoa and amoeba, bees were fed a formula of vinegar, citric acid, lactic acid, pollen and honey. The small number of hives that were infected with AFB and EFB were removed from the organic operation.
In conclusion, organic farming is still recommended over commercial beekeeping in monoculture farms, based on the cost related factors such as providing nutrition, transportation, labor, controlling diseases and loss of hives and queens.

[P.19.466] Complete wasps control in beehives without the use of chemicals and in a manner that supports the bees natural psychology

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Currently, apiarists are faced with the reality that Wasps, Mites and Beetles are ruining the productivity of hives and apiary operations. The damage to hives is most evident during non-nectar flow periods which usually coincides when the bees are weaker and more vulnerable to threats.
The research undertaken by Bee-IQ Solutions was done on the German wasp (Vespula Germanica), however, it is believed that HiveGateTM could have a positive impact on controlling additional invasive species. By observing what is happening inside beehives, during periods of wasp invasions, we began to understand the limitations that we have imposed on our commercial bees in creating an environment that unnecessarily exposes bees to being overcome by predatory wasps. The observations we made will challenge current wasp control practices.
HiveGateTM has been developed and serves the following functions:
1.Significantly reduces the number of wasps entering a hive.
2.Virtually eliminates any chance of wasps exiting a hive.
3.Is chemical free and enables the natural defence psychology of bees to be enabled.

4.In trials undertaken in high wasp areas, has given 100% protection of beehives and Nucs.
5.Allows for extended periods of hive protection without the need for frequent monitoring.
The primary mechanisms that have allowed HiveGateTM to be effective are based on the following:
1.Intruders are unable to enter the hive passively. Passive entry often goes unnoticed by the bees, HiveGateTM triggers alarm pheromones in intruders thus allowing bees to effectively respond.
2.Intruder entry is at the location of maximal bee defence – the cluster.
3.Intruder exit from a hive is extremely difficult as intruders continually avoid the exit point that is also the most defended point (the cluster). The continual release of intruder alarm pheromones inside the hive forces the bees to attack them.
4.The communication mechanism of intruders back to their nests is eliminated as wasps do not exit the hive.
5.Improved ventilation and humidity control is achieved by the vortex effect created by installing a HiveGateTM. The positioning is located at the highest source of humidity and heat generation, the bee cluster.
HiveGateTM has no moving parts, is cost-effective and long-lasting.

[P.19.467] Integrated varroa control in honey bee colonies and toxicity tests to caged adult varroa parasitized bees

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Maintenance of healthy honey bee (*Apis mellifera L.*) colonies is dependent upon the control of the parasitic mite Varroa destructor (varroa). Several attempts have been made to devlope resistance of honey bee colonies against varroa. Appropriate varroa diagnostic and control need to be performed in order to keep colonies productive. When using good beekeeping practices to maximize colony vigour, a beekeeper can combine appropriate diagnosis and applications of organic or combined control treatments which are normally non toxic to adult workers. We used either queen caging or brood removal to ensure adequate varroa control, after honey extraction or prior to the main spring honey flow. Integrated control methods were used at different times where none would offer sufficient control on their own. We studied comparative toxicity of three different solutions of OA dihydrate in sugar water, and several other acaricides: coumaphos, tau-fluvalinate, amitraz, thymol, and natural plant compounds (hop acids) on caged adult worker bees under laboratory conditions in to control varroa in colonies.
The highest cumulative varroa mortality during the 48h of exposure on caged bees was recorded in the Apivar® (98%) and CheckMite® (98%) treatment groups. The Apistan and HopGuard® both induced a 96% varroa mortality. The highest bee mortality was observed in the cages that were treated with HopGuard® and CheckMite® and the lowest bee mortalities occurred in the caged that were treated Apistan® 11% and Apivar® 7%. Apistan®, Apivar® strips, and Apiguard® gel were found relatively safe to adult bees.
In honey bee colonies we observed significant varroa mortality after total brood removal, or caging the queens and OA applications in broodless colonies, as well as in colonies with brood that received consecutive OA applications. Various acaricides are variably effective for varroa suppression when the mite populations are rising and brood is present. Hence, colonies that are treated with organic means in summer will require another treatment in autumn, so as to prevent a varroa resurgence. However, the control must be preceded by an accurate varroa diagnosis, in order to maintain the mite levels below an economic threshold that is needed for the colony remain productive.

[P.19.468] Effects of organic acids against Varroa destructor in Taiwan

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The varroa mite (Varroa destructor) is a major threat to worldwide apiculture. It was used to control the varroa mite by using tau-fluvalinate over 20 years in Taiwan. However the varroa mites have resistance to tau-fluvalinate has shown in Taiwanese apiaries recently. In this study, we evaluated the efficiency of varroa mite control by applying two organic acids, formic acid and oxalic acid. In autumn of 2016, we conducted different doses of slow releasing formic acid gel which was developed by ourselves, and also applied different doses of sublimated oxalic acid in autumn of 2018. The results showed that the single colony applied 25g formic acid gel had significantly higher varroa mites control efficiency 86% than treatment by tau-fluvalinate strips 73.2% ($P < 0.05$). That was also found in treated with oxalic acid in autumn of 2018. The single colony applied 2g oxalic acid had significantly higher varroa mites control efficiency 97% than treatment by tau-fluvalinate strips 84% ($P < 0.05$). These results suggest that formic acid and oxalic acid are great potential as alternative to against varroa mite in Taiwan.

[P.19.469] An anthropological approach to varroa control in France

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In France, the apicultural world, including scientists and beekeepers, professionals and amateurs, is schematically organized in two opposed poles. In one of them, scientists and beekeepers claim that the worst enemy for domestic bees in France are the pesticides and the European agricultural model. In the other pole, scientists and beekeepers claim that the worst enemy for domestic bees in France is

varroa destructor, this small mite arriving in France in 1982 from Asia.
In this paper, we will retrace the socio-history of a new field of techno-scientific and empirical knowledge about varroa which became, in less than 40 years, predominant in apidology. When varroa arrived in France in 1982, there was no knowledge about the interactions of this mite and the European bees. Everything was to discover. As a very well-known French biologist told us in an interview: "It was great at that time because we did not know anything! We had to discover everything!".
Since this time, scientists and beekeepers, in parallel and sometimes together, have tried to understand and to control the infestation of bees' colonies by varroa. Three different methods of varroa control coexist since the beginning: a genetic one, a chemical one, a biotechnical one. Each of them reveal different relationships to nature and knowledge and also some pragmatic elements according to the personal career (for scientists) and personal/familial/economic considerations for beekeepers.

[P.19.470] Building Better Beekeepers: Understanding Global Attitudes for Best Beekeeping Practices for Varroa, other Bee Diseases and Antimicrobial Resistance

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Bees are an intricate and complex species that require diligent care due to the numerous challenges to honey bee health. Pests, pathogens, poor nutrition, and pesticide exposure in concert with climate change are creating a difficult survival situation. Because humans depend so heavily on bees for both food and economic development, it is critical that the best beekeeping practices are identified and taught to the world at large.
In this study, we work to understand the global attitudes for best beekeeping practices when it comes to Varroa mites, other main honey bee diseases, and antimicrobial resistance. In the analysis of a series of surveys sent out in spring of 2019, we look to identify common beliefs and practices amongst beekeepers internationally. By having better insight into the way that beekeepers interact with their colonies, we will be able to help fill in knowledge gaps surrounding these important issues. Because research is constantly being done on bees and how to properly fight honey bee diseases and pests, it is pivotal that beekeepers around the world are in touch with the latest methods for keeping hives healthy and combating colony loss.
By learning more about how people interact with their bees, experts are able to give recommendations to beekeepers about how to improve bee management, survival rates and increase stability. This is extremely important especially in developing countries where economic growth for individuals depends upon bees health and their subsequent ability to produce goods. Learning more about the beekeeping audience and the ways that they think about hive management will allow for greater communication and connections between influencers and the greater community. By developing this network of beekeepers around the world, knowledge and information will flow more freely. Just like we depend upon each other for survival, so do bees; by working with our neighbors to address problems of bad practice, misinformation, and poor disease knowledge, we can create change in the beekeeping community.

[P.19.471] The liquid mixture of herbal extracts against Nosema apis and Nosema ceranae

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Nosemosis caused by Nosema apis and Nosema ceranae is a serious disease of honey bees *Apis mellifera L.* in worlwide and its prevalance is getting higher day by day. Since the fumagillin is forbidden in EU and most of the countries for treatment, some kinds of herbal tea become popular among beekeepers such as Thymus spp., Origanum spp., Salvia spp. etc. Beekeepers prepare the solutions by themselves without any consideration of doses, toxicity on bees, resistance of Nosema spores and level of Nosema infection. Nosemit® includes the extracts of Rumex acetosa, Achillea millefolium, Plantago lanceolata, Salvia officinalis, Thymus vulgaris, Rosmarinus officinalis, Laurus nobilis in liquid form. The aim of the study is to determine the efficacy of this herbal mixture against Nosema disease by cage and field experiments. The 50 honey bees were artificially infected with 200.000 spores/mL of N. apis and N. ceranae spore solutions in different cages. They were fed by 10 mL sugar syrup including 0,5 mL of Nosemit® . In the field studies, the number of Nosema spores were identified from the honey bee colonies of 5 different locations as the first step. Then Nosemit® 1/20 diluted by water was sprayed on colonies and frames. 15 mL of the solution was sprayed for each frame. The experiments were performed two weeks and Nosema spores were counted in every two days. The decrease of spore numbers is significant in both cages and hives. The lifespan of caged bees is also measured and compared with the control cages. The lifespan of experimental groups longer than the control groups and the difference is significant. Multiple ANOVA tests were used for all statistical analyses. The results from the field studies revealed that Nosemit® is more attractive to the colonies from the 3 locations where they have similar vejetation Nosemit® . Additionaly, Nosemit® is not affected by temperature via evoporation, because it is produced as liquid form. By this study, Nosemit® could be introduced as an alternative natural mixture against Nosema apis and Nosema ceranae for the near future.

[P.19.472] An Experimental Study on the Effect of an Unpredictable Incident on Colony Collapse Disorder (CCD)

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In beekeeping, Colony Collapse Disorder (CCD) is known to be a phenomenon that happens when the majority of worker bees disappear due to an extrinsic factor and leave the hive with the queen, food and a few nurse bees behind. The result of this decampment will be a remarkable decrease in the number of eggs that are laid by queen bee; which consequently leads up to a drastic reduction in worker bees population and finally the collapse of the colony. In some rare cases, the queen may leave the colony along with the worker bees in the time of disaster. A number of factors such as severe environmental changes, telecommunication waves and antennas, pathogens, pesticides, genetically modified crops such as transgenic maize, droughts, fungal agents, malnutrition, etc. have been studied and claimed to be the cause of colony collapse disorder over the past decades; although neither of them is absolutely proven nor rejected.
In this research, an experimental study was conducted in order to investigate the effect of a sudden unpredictable incident on colony collapse disorder by intentionally removing a considerable number of worker bees from the hive. It was observed that any alerting extrinsic factor that disturbs the queen bee can potentially lead up to this phenomenon .

[P.19.473] Four Decades of living with Varroa in Iran

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It was in 1978 that D.DE Jong, R.A.Mors and G.C Eickwort of the Department Entomology of the University Cornell in the USA published a title article: Mite pest of honey bees.Iran was' contaminated by Varroa mite
After four decades my experiences in France and Iran,also Afghanistan …etc.In the use of different types of treatments:by inhalation(fumigation),absorption(systemic action)and contact(slow release),evaporation against
of Varroa mite agree that the efficacy depends on the local conditions and that some care is needed.
In the Middle East ,Asia region,the optimal conditions are a high and stable external temperature and the absence of worker brood.The main precautions consist of avoiding reinfestation and robbing,in treating outside periods of nectar flow or queen rearing and in veriflying the efficacy of the treatment. The alternative to chemical control is thus more than the simple use of a vegetable or animal extract in place of synthetic acaricide.It requires an additional effort from the beekeeper to mange the bee colonies which is more time consuming.
Although we are in the 21 st century,Varroa destructor will undoubtedly remain for several years one of the principal agents of the weakening of apiarian livestock.Varroa destructor is a serious disease.
It is necessary to learn how to live with it.This can be done:
• By preserving only strang colonies in the apiaries
• By systematically changing the queen every two years,by developing queen selected for resistance to the diseases.
• The first treatment must be carried out in late September or early October in Iran and Afghanistan by chemical acaricide.To give over wintering bees the optimum potential for survival,it must be sufficiently effective to ensure that at the end of the treatment there will be fewer than 50 parasites within treated hives.
• If colonies were treated only with product of base in thymol and essential oils also formic-acid,they showed abnormally high winter losses,with clear evidence of mites.So it is necessary to use another acaricide chemical (Bayvarol®,ChekMite®,Oxuvar®…etc).
• If apiary's colonies are located in an area conductive to the reaing of brood(potential source of development for the parasite),the second treatment must be carried out in early sping.

[P.19.474] Microbial control of small hive beetle

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The small hive beetle (Aethina tumida) is native to sub-Saharan Africa but has spread and became a serious problem for beekeepers in North America. Traditionally, beekeeping pests have been controlled chemically. Today, a rapidly growing portfolio of microbial products expands pest management options in all areas of agriculture. Unlike chemicals, microbials are sustainable, biodegradable and not subject to resistance. Strong Microbials provide a novel pest control strategy: BioBeetle, a microbial pesticide which is toxic to small hive beetle larvae and repels adult small hive beetle from laying eggs in the hive.

[P.19.475] A survey of honeybee pathogens in Cuba

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Cuba is a tropical island country where beekeeping is an essential activity of agriculture. Cuban bees are of European origin, and since

the 1960s, bee imports have not been reported. Honey bee colony losses are estimated in 10% annualy. For cuban bees there is no information regarding the presence of different pest and pathogens, and their potential role in colony losses. Varroa destructor has been present in Cuba since the last century, but there are not information about its haplotype. In the present study, we analyzed 34 and 22 samples for the detection of Nosema spp. and RNA virus, respectively, from 11 provinces of the country. Haplotype determination of Varroa mites, we analyzed 160 adult female from 8 provinces. We detected Nosema ceranae, Acute bee paralisis virus (ABPV), Deformed wing virus (DWV) and Sacbrood bee virus (SBV) in Cuba. On the other side, Nosema apis and Black queen cell virus (BQCV) were not found. The detection of viruses in different provinces and the co-infection of colonies by several viruses and N. ceranae, indicates they are widely spread in the country. In the case of varroa destructor, all samples analyzed were of Korean haplotype. Since there are not studies about colony losses, consecuently we ca not say that the presence of these pathogens influences colony losses.

[P.19.476] Antimicrobial mechanism of two antimicrobial peptides produced by Bacillus against Paenibacillus larvae and Ascosphaera apis

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America foulbrood disease and chalk brood disease are microbial diseases caused by *Paenibacillus larvae* and Acsphaera apis respectively. At present, antibiotics are commonly used to treat them. Antibacterial peptides (AMPs) are a kind of biosafe bacteriostatic substances that have good inhibitory effects against bacteria, fungi and even viruses without causing microbial resistance. They are considered to be potential drugs to replace antibiotics. In this study, two antibacterial peptides, subpeptin JM4 and velezensin, were found from Bacillus subtilis and Bacillus velezensis respectively, which inhibited P.larvae and A.apis. The structure and function of subpeptin JM4 have been resolved, which are non-ribosomal peptides. Velezensin is a newly discovered AMPs with molecular weight of 4195 Da, while its structure and function need further study. Besides, the Antimicrobial mechanism of these two AMPs remains unclear. This study mainly focuses on the inhibition and antimicrobial mechanism of the two kinds of AMPs on P.larvae and A.apis. Firstly, the molecular structure and synthesis process of velezensin were identified by mass spectrometry, genome sequencing, heterologous expression and gene knockout experiments, and then in vivo and in vitro experiments were conducted to detect the inhibition of two kinds of AMPs on pathogens and their control effects on America foulbrood disease and chalk brood disease. Then, the effects of two kinds of AMPs on the cell membrane of pathogenic bacteria were detected. At last, the toxicity of two AMPs to bee larvae and adults was tested. It is expected to elucidate the mechanism and application potential of two new AMPs to control bee bacterial and fungal diseases, and to provide theoretical basis for the development of new drugs to replace antibiotics to control bee diseases.

[P.19.477] Development of Rapid and Sensitive Field Diagnostic Method of Sacbrood Virus

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Sacbrood virus (SBV) is one of the most serious honeybee viruses. The virus causes failure to pupate and death in both larvae and adult bees. Recently, Korean Sacbrood virus (KSBV) caused a great loss in Korean honeybee (*Apis cerana*) colonies. Therefore, a simple, sensitive and specific method for the field detection of SBV is needed urgently. In this study, a novel Ultra real-time PCR (GENECHECKER®) method was developed for rapid field detection of sacbrood virus (SBV) from honeybees (*Apis cerana*) infected with SBV in Korea. This novel Ultra real-time PCR system with a special polymer chip (Rapi:chip) allowing for fast thermal changes (8 °C/s ramping rate for both heating and cooling) and an integrated camera module for fluorescence detection, which is suitable for field diagnosis. In the field application of novel Ultra real-time PCR system, the whole diagnostic process was completed within 30 minutes including including reverse transcription reaction and total RNA extraction. In addition, this method could distinguish closely-related SBV strains. This molecular-based diagnostic tools was useful for the rapid and sensitive field diagnosis of SBV infection of honeybees.

[P.19.478] Long-term comparison of therapeutic efficacy of Fumagillin, prebiotics, and probiotics on Nosema infection, health and productivity of honey bee colonies

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Sustainable Canadian agriculture is in part dependent on a robust apiculture industry that provides healthy honey bee (*Apis mellifera*) colonies for crop pollination. Thus, recent unsustainably high colony losses have been raising concerns throughout Canada. Increasing evidence indicates that the microsporidian pathogen Nosema spp plays a significant role in mortality. Despite the significant negative impact of nosemosis on honey bee health, the only homologated treatment in Canada (i.e. Fumagillin-B) recently became unavailable due to discontinued production by the manufacturer. Moreover, the efficiency of commercially available alternatives have been often reported as contradictory. Accordingly, we initiated a long-term large-scale investigation to compare therapeutic efficacy of probiotics (i.e., Super

DFM and ProBee) and prebiotics (i.e., Nozevit and Honey-B-Healthy) to Fumagillin-B. A total of 105 experimental colonies were divided into 7 groups (n=15) and used in this experiment. 5 groups (total 75 colonies) were infected with Nosema spp. spores mixed with sugar syrup (~15,000 spores per bee) and subsequently treated with one of the above treatments according to manufacturers' instructions. In addition, one group (n=15) was challenged with Nosema but not treated (positive control) and another group (n=15) was not challenged and not treated (negative control). The variation between the spore loads found after inoculation (AI) and after treatment (AT) (δAI-AT) was used to assess the efficiency of the tested supplements. The ANOVA test showed a significant difference across all tested groups (F= 3.420, df= 5, P= 0.009) and the post hoc comparison showed a significant increase in the spore counts of ProBee treated colonies (X= 68.71, SD= 91.91) when compared to the untreated control colonies (9.48, 12.49) (Tuckey test: P= 0.027). Four weeks after treatment there was no significant effect of probiotic and prebiotic supplementation on the Nosema spore counts in experimental colonies. This experiment is in progress and evaluation of Nosema spore load, overwinter survival, and productivity of honey bee colonies exposed to above-stated treatments will be compared among experimental groups over two years.

[P.19.479] Comparing four methods of rearing Varroa destructor in vitro

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The parasitic mite Varroa destructor (Anderson and Truemann, hereafter Varroa) continues to devastate western honey bee (*Apis mellifera L.*) colonies throughout most of the world where they are kept. The development of a method to rear Varroa in vitro would allow for year-round Varroa research, rapidly advancing our progress towards controlling the mite. We created two separate experiments to address this objective. First, we determined which of four Varroa in vitro rearing methods yields the greatest number of offspring. Second, we attempted to improve the rearing rates achieved with that method. The four methods tested included 1) rearing Varroa on honey bee pupae in gelatin capsules, 2) group rearing Varroa on honey bee pupae in petri dishes, 3) providing Varroa a bee-derived diet, and 4) rearing Varroa on in vitro-reared honey bees. The number of reproducing females and the number of fully mature offspring were significantly higher in the gelatin capsules maintained at 75% RH than in any other method. A 3 × 2 full factorial design was used to test combinations of gelatin capsule size (7 and 6 mm diameters) and relative humidity (65, 75, or 85%) on Varroa rearing success. Varroa reproduction and survival were significantly higher in 7 mm diameter gelatin capsules maintained at 75% RH than those maintained in 6 mm capsules and at the other humidities. This work provides an important foundation for the development of future in vitro Varroa rearing protocols by identifying factors that influence Varroa reproductive success in vitro.

[P.19.480] Pollen fron Chilean native plants improve the survival of *Apis mellifera* infected with Deformed wing virus

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Honey bees (*Apis mellifera L.*) are key contributors to pollination of crops and wild plants worldwide. The beneficial influence of pollen nutrition on bee health is well-established. We therefore tested the influence of pollen diet quality (different monofloral pollens) and diversity (polyfloral pollen diet) on the survival of young honeybees infected with Deformed wing virus (DWV). Healthy honey bees (1 day-old) were confined in cages (60 bees per cage, 10 cages) and each cage was provisioned with pollen pellet (6 g) from (1) Chilean native Maytenus boaria (60 %), (2) Eucalyptus globulus (98 %), (3) Malus domestica (40%), Chilean native Maytenus boaria (20%), Chilean native Cryptocarya alba (20%), and (4) a mix of all them. A (5) pollen substitute was used as control treatment under the same controlled conditions (30 °C; 50-60% HR). Each cage was provided with sucrose (60% sucrose) syrup ad libitum. Five days later, one group of 5 cages for treatment were taken and the bees were infected orally with DWV (5 μL bee-1) and the other 5 were fed with saline solution. The survival (in DWV-infected and uninfected and fed with native and no-native pollen) of the honey bees was evaluated daily (by removing the dead bees in the cages) during 20 days. At the end of the assay, in treatments not infected with DWV, honeybee survival was circa to 50%, except in the treatment in which the bees were fed with pollen of Eucaliytus globulus (26%). On the other hand, in the treatments infected with DWV, the highest survival was obtained with polyfloral pollen (59%), with an increase in survival over 90% compared to the control (30%); followed by the pollen mixture, with a survival of 56% and the monofloral Maytenus boaria pollen with 46%. Given these results we can conclude that the pollen from Chilean native plants improves the survival of the infected-DWV bees.

[P.19.481] Rapid identification of Paenibacillus larvae using matrix-assisted laser desorption ionization-time of flight mass spectrometry

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American Foulbrood (AFB) disease caused by the spore-forming bacterium *Paenibacillus larvae* is a highly contagious disease affecting the larval and pupal stages of honey bees (*Apis mellifera L.*). AFB is the most widespread and destructive of the brood diseases, and possesses unique problems for prevention and control because the spores can remain viable for long periods and survive environmental adversities. We used matrix-assisted laser desorption ionization-time of flight mass spectrometry (MALDI-TOF-MS) for rapid identification of these pathogenic bacteria. MALDI-TOF-MS is a rapid, precise, and cost-effective method for identification of bacteria, compared to conventional identification techniques, such as biochemical and molecular biological tests. Identification by MS analyzes the profiles of bacterial macromolecules that are obtained from whole bacteria. This novel proteomic approach allows rapid and accurate identification of bacteria as well as yeasts and fungi. In this study, 46 isolates were tested in duplicate by MS analysis; 21 of 46 isolates (45.7%) were identified as P. larvae by MS analysis, which were confirmed as P. larvae by 16S rRNA genetic identification. Compared to the standard strain, the isolates were identified as P. larvae by MALDI-TOF-MS, which shared major peaks but showed different intensity, compared with P. larvae ATCC 9545 as reference strain. In addition, MALDI-TOF-MS could distinguish pure culture isolate of P. larvae from mixed cultures containing other bacteria. In the future, it is needed to accumulate more and more MALDI-TOF-MS profiles of P. larvae for the improvement of identification accuracy by MS analysis. If so, MALDI-TOF-MS will be a useful and rapid technique of P. larvae identification replacing the conventional identification methods.

[P.19.482] Evaluation of two methods of treatment with oxalic acid for the control of varrroa destructor in honey bee colonies *Apis mellifera L.*

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The parasitosis caused by the mite Varroa destructor is considered one of the diseases with the greatest impact on apiculture worldwide. The objective of the present work was to evaluate the effectiveness of two treatment methods based on oxalic acid, one is the liquid application in combination with glycerin and the other applied in the form of gas by the sublimate method, for varroa control. The work was carried out in an apiary located in an avocado orchard, located in the municipality of Tancitaro, Michoacán, Mexico. We worked with langstroth hives with colonies of the hybrid of *Apis mellifera L.*, The hives were divided in three groups for each type of treatment: Treatment one: Oxalic acid in combination with liquid glycerin, Treatment two: Application of pure oxalic acid by sublimate method and Treatment three: group of control beehives. The effect of the treatment on the size of the nest of the colony was also evaluated. It was found that the use of oxalic acid in both forms of treatment is effective for the control of varroa in the phoretic stage without significant differences with an 87.89% on average, however the sublimated treatment generates a lower fall of mites (5.4 per day), even compared to the control (6.8 per day). Regarding the influence of treatments on the condition of the hive, it was observed that there is a decrease in the size of the nest in the use of sublimed oxalic acid (-25%) compared with the treatment of oxalic acid in combination with glycerin (+ 2.5%). Oxalic acid is considered as an alternative commonly used among mexican beekeepers for the control of varroa, although it is effective for the control of the infestation by at least 80%, but depending on the method used this can cause adverse effects in the condition of the colony, from the evasion to the mortality of the population, as well as the decrease in the size of the nest, It´s necessary more research oriented for evaluating different methods and dose of treatment, as a strategy for mite control and exposure to different climatic and production conditions.

[P.19.483] Control of Varroa destructor by oxalic acid in cardboard strip

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The ectoparasitic mite Varroa destructor is the main honey bees´ health problem in the world. Given the growing resistance of bees to synthetic acaricides, the use of organic acaricides has been revalued. Oxalic acid is an organic acaricide widely used with variable success due to the difficulties of application. In Uruguay, because the colonies must receive acaricides to survive during the winter, beekeepers began to use a formulation of oxalic acid and glycerin in cardboard strips. The aim of this project was to measure the effectiveness of this product to control V. destructor and determine if it has negative effects on the population of bees. For this, in autumn to 11 beehives this acaricide was applied according to its population (one strip every 2.5 frames with bees) and 10 colonies were assigned as control. Treated colonies and controls had the same population of adult bees (6.3 ± 1.4 and 6.9 ± 1.9 frames covered with bees, respectively) and breeding (0.6 ± 0.6 and 0.6 ± 0.4 honeycomb faces, respectively); and similar levels of V. destructor infection (3.4 ± 2.4% and 2.4 ± 2.0%, respectively) when applying the acaricide. The product was left for 41 days during every 5-7 days fallen mites were collected. Dead bee traps were also placed in three hives of each group. When the strips were removed, amitráz and fluvalinate were applied to this hives and the mites that survived the oxalic acid were collected. The efficacy of the oxalic strips was 95.3 ± 3.6%. The percentage of mites that fell after the application of oxalic acid was 55% at 8 days, 77% at 14 days and 93% at 22 days. Three of the colonies that did not receive oxalic acid collapsed while the remaining 7 increased the infestation by V. destructor by 67%. No differences were observed in the number of bees collected in traps of dead bees between the treated and untreated colonies. The results show that cardboard strips impregnated with oxalic acid and glycerin are effective in the control of varroosis.

[P.19.484] Transmission of some pathogens between honeybees and Bumblebees

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The pollination benefits of honeybees and wild pollinators in food production and biodiversity protection is well known so it is extremely

important to recognise the threatening factors and possibility for introduction of appropriate measures.
During the year 2017 and 2018, the research on some pathogens of *Bumblebee*s in comparison to honeybees was done in Slovenia. In total, 132 *Bumblebee* workers were collected in nature at four locations. At the same time and the same location, also honeybees were sampled. In the laboratory, the occurrence of deformed wing virus (DWV), chronic bee paralysis virus (CBPV), acute bee paralysis virus (ABPV), black queen cell virus (BQCV), sacbrood bee virus (SBV) and Lake Sinai virus (LSV) were detected by using the RT-PCR methods. The presence of Nosema bombi, Nosema ceranae, Nosema apis, Apicystis bombi, Crithidia bombi, Crithidia mellificae and *Lotmaria passim* were identified by using the specific PCR methods. In both *Bumblebee*s and honeybees, the presence of five different viruses and other above listed pathogens were confirmed, with differences according to *Bumblebee* species, location and year of sampling. These results indicate the possible transmission of some pathogens between honeybee and wild pollinators. To find out how these pathogens are transmitted within the *Bumblebee* population, in the early spring 15 *Bumblebee* queens from the same locations were also included in the investigation, as the only ones that overwinter and form a new colony in the season. In the above described laboratory tests, only the presence of BQCV, Apicystis bombi and Crithidia bombi was found in *Bumblebee* queens while tests for all other pathogens were negative. We can assume that except BQCV the *Bumblebee* queens does not transmit honeybee viruses and Nosema sp., thus *Bumblebee* workers could be infected in some other way.
In further research, the possibility of disease transmission between honeybee and different wild pollinators during their pollination activity should be investigated. This is very important, because if some pathogens are transmitted in this way between honeybees and *Bumblebee*s, they can also be transmitted between honeybees, especially at high density of bee colonies in the areas with rich honey flow.

[P.19.485] Effects of an extended release oxalic acid treatment on Varroa destructor mites and *Apis mellifera* honey bee colonies in the US Southeast

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The introduced ectoparasitic mite Varroa destructor is among the most significant of biological stressors to *Apis mellifera* honey bees. The mite vectors viruses and feeds on host fat body; parasitism can result in multiple negative effects including shortened lifespan of individual honey bees and ultimately colony death. To control V. destructor, beekeepers often rely on a combination of acaricides. Oxalic acid is one such organic compound that shows considerable promise. Because acaricidal effects of oxalic acid do not penetrate wax brood cell cappings, there is an interest to identify a novel extended release technique that targets V. destructor emerging from brood cells alongside their *A. mellifera* hosts. To study this, we established 20 double-deep brood chambered colonies in an experimental apiary in Alabama; half the colonies each received 1.5 shop towels that were previously treated with 18 g oxalic acid diluted in glycerin and water (1:1:1 weight ratios); the other half received no shop towels. Colonies were assessed weekly for V. destructor intensity, and every three weeks for *A. mellifera* worker populations and colony weight. Effects of this extended release oxalic acid technique on V. destructor and *A. mellifera* colonies will be discussed, as will considerations for its use in apiculture.

[P.19.486] Evaluation of vegetal extracts and metabolites obtained from winter`s bark (Drymis winteri) as potential veterinary product for control and treatment of varroosis in honey bees

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One of most important problems presents in Chilean beekeeping is Varroosis, parasitary disease caused by mite Varroa destructor. In fact, prevalence of Varroosis has been estimated at 72% by epidemiological surveillance performed by sanitary authority (SAG). This disease is characterized by mites externally digest and consume fat body tissue. The application of synthetic acaricides has been conventional method to control Varroosis, however in last decades numerous studies have reported the generation of resistance. Development of natural acaricides for control and treatment of V. destructor may reduce use of synthetic products in beekeeping. Essential oils (EO) have demonstrated optimal results to control and treat Varroosis if low infestation levels are diagnosed in honey bees colonies. In this context, EOs correspond to highly volatile liquid compounds that would maintain infestation rates under economic damage threshold for beekeepers. Due to the above, it is extremely urgent to find new treatments to cope with this disease, which hopefully are harmless for both people and the environment. Canelo (Drymis winteri) a Chilean endemic tree, emerges as an interesting alternative to evaluate, since it has been demonstrated contains terpenes of drimane type and flavonoids. In plants, drimanes fulfill several functions, nevertheless until now their effects on survival of adult Varroa in vitro has not been investigated. In this work, chemical properties of semipolar extracts and of the active principles (drimenol and polygodial) obtained from D. winteri from five Chilean southern locations were assessed. These semipolar extracts were characterized by high performance liquid chromatography (HPLC). The HPLC pattern of these metabolites varies according to origin place of D. winteri´s bark, which allowed to quantify a concentrations range of 0 and 48197 parts per million (ppm) of drimenol. Toxicity tests of vegetal extracts demonstrated that they were not toxic for young honey bees (survival rate ranges between 53% and 93% after 6 hours exposure) maintained in vitro. More recently, we have isolated drimenol and polygodial from these vegetal extracts through chromatography in column (TLC), and characterized them by NMR spectroscopy. These experiments may provide new acaricide molecules for control and treatment of Varroosis in honey bees

[P.19.487] Four years of Field Trials Conducted in Saskatchewan for the Determination of Formic Pro™ Efficacy Against Varroa destructor, effects on Deformed Wing Virus and Nosema Levels in Colonies of Honey Bees

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The objective of this series of Formic Pro™ (MAQS+) field trials was to determine the efficacy and impact of applying the formic acid vapour delivery technology at different time periods (early spring, late spring, late summer and early fall) in Saskatchewan, Canada. The effects on phoretic varroa mite levels, varroa viability in sealed brood, DWV virus profiles before and after treatment, nosema levels and queen status were determined. Eight trials were performed over 4 years involving 270 colonies. The temperature conditions were monitored for all trials to assess the effects of the temperature conditions on efficacy and queen mortality. The treatment periods were also varied (7 to 14 days). Live and dead adult worker and drone bees were collected before and after formic treatments to determine DWV levels and distribution between different castes at different times during the season. Composite samples of bees were extracted from selected trials and subjected to QPCR to determine the colony virus levels. Nosema levels were also determined by PCR analyses after formic treatment. Phoretic mite levels were determined by alcohol washes of at least 300 adult bees and brood infestation levels and varroa viability assays were assessed visually by opening ~100 cells. In general, these trials of the Formic Pro™ technology showed good efficacy with little effect on colony activities other than some short term effect on brood production. Temperature had an effect on queen mortality, with higher mortalities at higher temperatures, but better varroa kills. The 14-day treatment period trials showed the best results with good to excellent efficacy and the highest levels of varroa kill in sealed brood.

[P.19.488] Beneficial microbes as a strategy to manage nosemosis

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The administration of beneficial microbes (BM) as food additives is a widely used strategy, both in humans and animals, to improve health. In previous studies, our research group obtained a mixture of BM composed of 4 strains of Lactobacillus kunkeei, isolated from the intestinal microbiota of bees. This mixture was able to decrease the mortality due to *Paenibacillus larvae* infection, the causative agent of American Foulbrood, in larvae.

In this study, we aimed to evaluate the in vivo effect of the administration of these BM against Nosema ceranae. This microsporidia exerts a negative impact on honey bee health, affecting host cell cycle, immunity and apoptosis.

New emerging bees *Apis mellifera iberiensis* were confined into cages and fed ad libitum with a sugar syrup solution supplemented with Promotor L. Two days later, two groups of bees were fed ad libitum with the BM in sugar syrup (final concentrations of 1x108 ufc/ml, groups BM and BMN) while two groups only received sugar syrup (C and N). Seven days after emergence two of these groups (BMN and N) were fed individually with 1x105 N. ceranae spores, while the others received sugar syrup (BM and C). The assay was carried out in triplicate. Bees were taken out of the incubator daily recording mortality. Samples were taken 4, 7 and 12 days post infection, to evaluate the expression levels of genes involved in host cell cycle, immunity and apoptosis, by RT-qPCR. The development of N. ceranae was also evaluated by RT-qPCR using the expression of ptp3 as a marker.

These BM were able to decrease the number of N. ceranae spores, although no difference was observed in the lifespan of the infected bees. Besides that, BM were able to alter the expression level of different genes, in some cases to counteract the immunosuppression caused by N. ceranae. These results encourage the development of new studies using these BM to improve bee health.

[P.19.489] Efficacy of Api-Bioxal and ApiHerb against Nosema ceranae investigated by two q-PCR methods

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Nosema ceranae is an emerging *Apis mellifera* pathogen parasitizing the epithelial cells of honey bee ventriculum. In the past years, it has become one of the most prevalent honey bee pathogens globally. The infections, known as Nosemosis type C, are detrimental both at the individual and at the colony level, making N. ceranae one of the major drivers of colony losses. Legal restrictions and poor availability of effective active ingredients make options for treatment limited, especially for the organic beekeepers.

Based on the results of preliminary tests, the formulations Api-Bioxal and ApiHerb were tested comparatively in the field for their efficacy against N. ceranae infections. The products are an oxalic acid-containing antivarroa miticide and a feed supplement based on plant extracts, respectively. Both were used as per the label instructions by trickling, after dissolving them into sugar water (1:1 w/v).

In October 2017, eighteen colonies were selected from a naturally infected apiary located in Bologna, Italy, and randomly split into

three treatment groups (N=6), one of which served as the untreated control. The other groups received respectively one Api-Bioxal administration, and three ApiHerb administrations one week apart.
Twenty-five old workers were sampled from each colony pre- and post-treatment. q-PCR was used to measure N. ceranae abundance in the individual bees through sequences of the Hsp70 and 16S rRNA genes.
The Hsp70 method resulted in significantly lower abundance. This is compatible with the nature of the 16S rRNA gene, which may be present in multiple copies in the N. ceranae genome. On average, the treated groups exhibited a decreased N. ceranae abundance, which was not detected in the untreated colonies. The reduction ranged three to four orders of magnitude, depending on the q-PCR method that was used. Besides, at the post-treatment sampling, some of the ApiHerb treated colonies failed to show infected by N. ceranae.
The results above are in line with previous findings on oxalic acid and ApiHerb. If confirmed by further trials, they may indicate a way to control N. ceranae infections by an environmentally sound approach.

[P.19.490] Understanding the differential hygienic behavior towards drone brood in *Apis mellifera* colonies from Argentina

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Brood diseases of *Apis mellifera* colonies constitute a main problem of beekeeping worldwide. Worker bees display a social health mechanism that consists in detecting, uncapping and removing dead or diseased brood from the hive: the hygienic behavior (CH). These activities are induced by olfactory cues and have been described as associated to hygiene of brood parasitized by Varroa destructor. This mite have preference for drone brood, but the efficiency of CH towards their cells is significantly lower compared with cells of worker brood, being left uninspected by workers. Some authors suggest that a possible cause of the CH differences is due to the cell wax cap of drone brood (thicker than worker cells) acting as a barrier to volatile compounds and obstructing disease detection. The aims of this research were to study the differential CH towards worker and drone brood belonging to highly hygienic colonies from Argentina, and to explore the importance of drone cell wax cap as an interfering factor in the transmission of chemical signals. To this end, removal percentages of pin-killed worker and drone brood were recorded and an innovative cell wax cap exchange was implemented in three different treatments: pin-killed worker pupa with a healthy drone cell wax cap; a healthy worker pupa with a pin-killed drone cell wax cap; and a healthy worker pupa covered with a healthy drone cell wax cap (control). Results showed a greater removal towards worker cells than drone cells. For the cell wax cap exchange experiment, we found that the removal of pin-killed worker pupae covered with healthy drone cell wax cap was significantly high, while the removal of healthy worker pupae covered with pin-killed drone opercula was low. These preliminary results confirms a differential behavior between both type of brood cells and suggests that the cell wax cap of drone brood is not interfering the detection of chemical compounds from the diseased brood by worker bees, regardless the thickness. This work contributes to a better understanding of the detection activity of different types of diseased brood and provides information useful to control strategies of varroosis and other brood diseases.

[P.19.491] Formic acid treatment against Varroa destructor in *Apis mellifera* colonies: Effects on mite mortality and brood

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Formic acid is widely used to control Varroa mites (Varroa destructor) in conventional and organic beekeeping. Despite its intensive use, many key parameters of this active ingredient are still unclear, including its pharmacodynamics and the effect of environmental conditions on treatment efficacy. Inefficient treatments may lead to colony losses and a thorough understanding of its mode of action under field conditions is therefore required.
We tested commercially available dispenser systems differing in the amount of formic acid applied and in the duration of formic acid evaporation in the hives at different weather conditions. Our results show, that long-term application of high amounts of formic acid (average of 406 g formic acid for 12 days in a hive with a volume of 75 L) showed the highest efficacy, but also affected the brood and the brood rearing activity in the colonies at elevated air temperature, even though it had no long-term effect on colony survival and colony strength in spring. We could demonstrate that formic acid affects also mites in capped brood cells, but this effect was small. Ambient air temperature and relative humidity had no effect on formic acid evaporation in the hive.

[P.19.492] Semiochemical composition of the mandibular, postpharyngeal glands and head cuticle of Vespa velutina by gas chromatography-mass spectrometry (GC-MS)

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Hornets are classified as eusocial species, with colonies composed of thousands of individuals, where chemical communication plays an important role in their lives (Keller and Nonacs, 1993). These chemicals are called semiochemicals and the best representative ones are pheromones. Cuticle components, especially hydrocarbons, are a good example of this action, playing a role in communication between individuals of the same species and perform nest mate recognition functions.

Vespa velutina is a hornet recently introduced in Europe, reaching the Iberian Peninsula in 2010. This invasive species is causing a great socio-economic and environmental impact related to its enormous capacity for proliferation, spread and adaptation. The beekeeping sector is one of the most damaged since honeybee is part of its diet. Spring bait-trapping is one of the main control methods but nevertheless is a non-specific method, seriously affecting biodiversity (Rodríguez-Flores et al., 2018. The improvement of specific attractant pheromones in the bait-trapping method can be a useful tool to minimize this impact.

The aim of this work was the extraction of the semiochemicals of V. velutina heads and its chemical identification by gas chromatography-mass spectrometry (GC-MS).

Fifty-two compounds were identified from the mandibular and postpharyngeal gland's secretions and from the head cuticle. Among the compounds with the highest concentrations (> 250.0 ng/mL) it was possible to identify L-(-)-sorbofuranose and D-(-)-tagatofuranose, sugars which are molecular energy-providing nutrient and a basic substrate for the biosynthesis of many macromolecules. As the main hydrocarbon, probably from the cuticle, octacosane was detected. Finally, oleic acid, palmitic acid and stearic acid were the major long-chain carboxylic acids from the secretion of the glands of V. velutina.

Keller, L., & Nonacs, P. (1993). The role of queen pheromones in social insects: queen control or queen signal? Animal Behaviour, 45(4), 787-794.

Rodríguez-Flores, M. S., Seijo-Rodríguez, A., Escuredo, O., & del Carmen Seijo-Coello, M. (2018). Spreading of Vespa velutina in northwestern Spain: influence of elevation and meteorological factors and effect of bait trapping on target and non-target living organisms. Journal of Pest Science, 1-9.

[P.19.493] Evaluating brood mixing as a method of inducing simulated hyperpolyandry for increased colony resilience

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Intrinsic resilience in *Apis mellifera* to stressors such as Varroa destructor has been increased by creating hyperpolyandrous colonies via queen artificial insemination. We evaluated a low-tech method of delivering benefits of polyandry – simulating a hyperpolyandrous state by mixing sealed brood within sets of colonies. Frames of closed brood were shared equitably within designated colony sets; performance of these colonies was compared against those in unmixed sets. Brood mixing created a temporary surge in patriline number per colony and was expected to mirror benefits of having a single hyperpolyandrous queen: namely the capture of rare specialist alleles that are already present within a population but are unable to be sufficiently expressed in a single colony due to natural constraints to mating number. The temporal longevity of this beneficial effect was extended with subsequent remixing amongst treatment colonies. Worker population, brood area, and mite counts were regularly assessed to compare mixed and unmixed colonies. Considerations for applied practice and associated risks are presented.

[P.19.494] River Disease in honebees: clinical presentation and management

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The River Disease (RD) is a clinical presentation caractherized by massive death of larvae of bees, produced by the secretions of the insect Epormenis cestri associated with Sebastiana scottiana trees. The affected colonies usually die due to depopulation or become unviable for wintering. These trees are found on the coast of important water courses. The secretions are produced from November to February (austral spring-summer). The clinical presentation can be classified according to whether the death of larvae is total (severe), or partial (moderate or mild). The apiary is completely affected, and the most notorious clinical signs are the absence of larvae (severe), or the spotty brood pattern (moderate or mild). In advanced stages the progressive accumulation of pollen in the breeding chamber, which can occupy several honeycombs, and the presence of cells with abundant royal jelly, without larvae are the main characteristics. The adult bees do not seems to be affected, and they continue to work normally, until they begin to depopulate. The managements that have been evaluated as simpler and more effective, are oriented towards the removal of toxic honeydew and the contribution of some combs from unborn bees from healthy colonies. The most common situations are: 1) removal of colonies from the RD zone. If performed early (first clinical signs), the colonies usually recover without any management, as long as there is a good flow of nectar in the new area where they are located. When they are removed in more advanced stages, it should be completely harvest the hives, provide 1 or 2 combs of unborn bees and in the case that there is no entry of néctar you must provide sucrose syrup since the colonies are left without reserves; 2) the colonies are kept in the RD zone, be provided 1 comb of unborn bees every 3 weeks and only when cessation of Epormenis cestri secretions should be completely harvested. In both situations the development of breeding and the population of adult bees should be monitored in order to ensure that they achieve a good population of bees for wintering.

[P.19.495] Effect of artificial media and temperature on the growth bee brood pathogen Ascosphaera apis and optimization its cultivation in vitro

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Ascosphaera apis is a causative agents of chalkbrood, which belongs to the most widespread bee brood diseases. It can significantly reduce bee numbers and colony productivity, especially in humid and stress-related areas. It is closely adapted to bee brood and is marked by many differences from other insects pathogenic fungi, particularly its only way of sexually reproduction. For this reason it is not possible to use many well-known cultivation methods for common entomopathogenic fungi. In this paper there were compared influence of several artificial media and cultivation under different temperatures on the growth and development of both, female (+) and male (-) mating types of local strains of A. apis. There were carried out 3 experiments to better evaluation of suitable conditions for local strains. In the first experiment, radial growth of separated mating types was measured daily. In the second one spore cysts, spore balls and ascospores were counted simultaneously in a Neubauer haemacytometer chamber. This technique does not allowed counting real number of spores, but it suits very well for assessment of A. apis development characteristics in different conditions. The last experiment was focused on a morphometry of spore cysts and spore balls. There was found a correlation pattern between reproductive structures size and temperatures, which was very useful for overall assessment. As a result of these experiments, the best temperature for in vitro cultivation both, male and female local mating types, was determined as a 30°C. SDA and YGPSA media are suitable for fast growth. For reproductive structures production are the most useful media PDA-BB4, which was newly designed for this purpose, and SDA. In the most countries (European Union, Australia), there is not any registered therapeutic agent against chalkbrood therefore it is important to understand life cycle and pathogenesis of Ascosphaera apis, but especially, to optimize cultivation methods for better observations in vitro, which was main reason of this study.

[P.19.496] Sharing the experience about the control techniques of harmful hornets to honey bee colonies

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Some Vespa hornets can cause many damages to honey bee, *Apis mellifera*, colonies. In North Africa and the Middle East, Vespa orientalis or commonly known as the oriental hornets are common and harmful to some fruit crops and honey bee colonies. The oriental hornets can attack forager workers, drones, and virgin queens during their flight activity. Also, these hornets can invade and destroy bee colonies. Recently, the Asian hornets, Vespa velutina, have invaded Europe and caused damages to beekeeping sector. In this poster, I present the techniques used in the Arabian countries to control the oriental hornets. This can help researchers and beekeepers in other countries to control endemic and exotic hornets and to develop new control techniques in light of the presented methods. For the oriental hornets, the control strategies concentrate on controlling queens during spring, destroying nests during summer, and trapping hornets during autumn. The techniques used to realize these strategies are presented as well as various designs to hornet traps. Moreover, new trends to control hornets are presented at the end of the poster including the utilization of the geographical information system (GIS), remote sensing (RS) and harmonic radar to control hornets

[P.19.497] Investigating the fumigating toxicity of Satureja khuzistanica essential oil on the mortality of varroa mite (Varroa destractur) and *Apis mellifera meda*

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Varroa mite (Varroa destractur) is one of the most important pests of honey bee colonies in different parts of the world including Iran. The mite feeds on the larvae hemolymph of honey bees and causes the spread of viruses resulting in considerable damage to the industry of honey bees, worldwide. In the recent years, and due to the increased resistance of varroa mites to acaricide, the mites contaminate the honey bee products including bee, wax and Royal jelly. Accordingly, in this research, it was hypothesized that chemicals, which are of natural origin, such as the essential oils of Satureja khuzistanica, with fumigating properties (less harmful for human), are suitable replacement for miticides controlling varroa mites in the colonies of honey bees. The objective was to investigate the toxicity of Satureja khuzistanica essential oil on the mortality of varroa mite (Varroa destractur) and *Apis mellifera* meda. In the lab preliminary experiments,

with the temperature of 30±2 oC and the relative humidity of 50±5 (%), varroa mite and *Apis mellifera* meda were treated with the essential oil (40-300 mg/kg). The mites and honey bees were treated in Petri dishes, with the length of 10 cm and the height of 3 cm, and with a filter paper at the bottom. The preliminary and final experiments were factorial on the basis of completely randomized block design with four replicates. The rate of carvacrol natural product (with acaricide property), present in the essential oil, was 93%. The LC50 of essential oil (after 10 h) for the mite was equal to 78.55 mg/kg with the confidence interval of 31.58-195.32, and for the honey bee was equal to 535.78 (325.11-880.42). This indicates that the essential oil of Satureja khuzistanica results in the least rate of mortality and toxicity for honey bees, and has a high potential for controlling the mite in the colonies of honey bees. It is also possible to enhance the effectiveness of the method, if the essential oil concentration and fumigation time is adjusted accordingly.

[P.19.498] Study of the larvicide effect of toxins of two local strains of Bacillus sp. against the greater wax moth Galleria mellonella

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To reduce the dependence of the agricultural sector on chemical pesticides, the use of biopesticides is proving more and more effective and recommended, the formulation of biocides based on micro-organisms becomes more and more efficient.
Our work is based on the exploitation of the larvicidal potential of two bacterial strains of genus Bacillus; B11 and B8 by their use against the larvae (L5) of the wax moth *Galleria mellonella*. Their effect on the haemolymphatic metabolites (proteins, lipids and carbohydrate) of the larvae has also been studied.
For this, a rearing of the insect took place under controlled conditions. The bacteria were isolated from the rhizosphere of a cultivated soil in the Boumerdes region (located in northern Algeria), their characterization and taxonomic identification took place based on their morphology and their physiological and biochemical characteristics. In addition, a molecular study was carried out by sequencing of 16s RNA with phylogenetic sequence analysis. This study allowed us to identify both strains as being; Bacillus mycoides strain 11 and Bacillus clausii strain B8.
The acid and alkaline extraction of toxins from both bacteria was followed by a study of the toxicity of these secondary metabolites to the insect. For this three doses were used: D1 = 15 µl / ml, D2 = 30 µl / ml and D3 = 60 µl / ml, the controls are treated with sterile distilled water. In the light of the results obtained, it is found that the toxins extracted from the two bacterial strains B11 and B8 have a significant effect on *Galleria mellonella* L5 larvae. They caused very remarkable symptoms and behaviors such as a strong decrease of appetite and malformations with mortality rates which vary according to the dose. On the other hand, the determination of hemolymphatic metabolites revealed an increase in protein and carbohydrate levels and a decrease in lipid levels.

[P.19.499] Sensitivity / Resistance dynamic of the mite, Varroa destructor, to the acaricides : contribution to integrated pest management

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Varroa destructor is an ectoparasite mite of *Apis mellifera*, responsible for varroosis. Without treatment, the honeybee colony collapse in few years. Only three active substances are commonly used as chemical acaricides in France: flumethrin, tau-fluvalinate (both pyrethroid) and amitraz. After many years of use, the mite developed some resistance to these active substances, which can lead to reduced efficacy. The tau-fluvalinate resistance was studied by many researchers and the genetic resistance mechanism starts to be well-known. Specially one mutation can explain the resistance mechanism of the mites. However, for now, the amitraz resistance is not understood even for uses in agricultural. One of the resistance hypothesis to amitraz is a metabolic resistance. Yet, in some countries, amitraz resistance has been proved by lab test but not by genetic test. In France, this lab test is developed to evaluate the amitraz and tau-fluvalinate resistant population. To evaluate a resistant population, a referent "sensitive" population is needed. In 2018, referent and resistant populations of varroa for amitraz have been discovered. In 2019, the same work will be realized for tau-fluvalinate and carry on for amitraz. Many collaborations will allow us to find resistance centers and describe the situation of resistance in France. If these centers are discovered some measures could be applied to limit the dispersion of the resistance. The main objective of this work is to develop a method to evaluate the sensitivity of the varroa mite to the active substance to advise beekeepers in their choice of acaricide and limit the development of the resistance.

[P.19.500] Disease Resistance through Enhancing the Queen Nutrition

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Arami Bokan, Bokan, IRAN

The nutrition level of bees determines their susceptibility to disease. Improving bees' nutrition by moving colonies to protein rich areas, selecting for bees with superior foraging behavior, or using nutritional supplements that cater to bees biological needs will greatly ameliorate

a colony's chances of withstanding biological disease.
Colonies with efficient forages improve a hive's nutrition. Selecting for foraging behavior leads to stronger immune systems and hygienic behavior. Therefor selection and nutrition are key to colonies improvement.
In our research, we have found queen larvae is mostly unaffected by American Foulbrood (AFB), European Foulbrood (EFB), Chalkbrood and Sacbrood, and we have concurred this is due to the queen being feed royal jelly. Observation made in Canada and Iran of hundreds of queenless hives with active AFB found that bees can select infected larvae and still produce disease- free queens. We concluded that royal jelly impart disease resistance and protect against certain pathogens. Thus, royal jelly is important to any bee disease control regiment.
For the past seventeen years, we have been using genetic selection and increasing the nutrition of bees with Caspian Solution, a blend of royal jelly, pheromones, and other natural ingredients.
Key words: royal jelly, Caspian Solution, American Foulbrood, selecting, nutrition

[P.19.501] The EU-funded B-GOOD project: Giving Beekeeping Guidance by cOmputatiOnal-assisted Decision making

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A key to healthy beekeeping is the Health Status Index (HSI), inspired by the European Food Safety Authority's (EFSA) Healthy-B toolbox. The recently EU-funded B-GOOD project aims to make the Healthy-B toolbox fully operational through the active collaboration of beekeepers by facilitating the coordinated and harmonized flow of data from various sources (hive, bees, beekeeper) through to colony health and productivity. B-GOOD envisages a step-by-step expansion of participating apiaries to eventually cover all EU biogeographic regions. The key to sustainable beekeeping is a better understanding of its socio-economics, particularly within local value chains, its relationship with bee health, and the human-ecosystem equilibrium of the beekeeping sector. B-GOOD aims to implement these insights into data processing and decision making. B-GOOD will fully integrate socio-economic analyses, identify viable business models tailored to different contexts for European beekeeping, and determine the carrying capacity of the landscape. In close cooperation with the EU Bee Partnership, comprising a mix of institutes, scientists and major stakeholders, an EU-wide bee health and management data platform and affiliated project website will be created to enable sharing of knowledge and learning between scientists and stakeholders within and outside of B-GOOD. We will utilize and further expand the classification of the open source IT-application for digital beekeeping, BEEP, to streamline the flow of data related to beekeeping management, the beehive and its environment (landscape, agricultural practices, weather and climate) from various sources. The dynamic bee health and management data platform will allow us to identify correlative relationships among factors impacting a Health Status Index (HSI), assess the risk of emerging pests and predators, and enable beekeepers to develop adaptive management strategies that account for local and EU-wide issues. Reinforcing and establishing, where necessary, new multi-actor networks of collaboration will engender a lasting learning and innovation system to ensure social-ecological resilient and sustainable beekeeping.

[P.19.502] Sharing is caring: Validation of the Varroa alert system, a freely accessible, web based data sharing platform for beekeepers

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Coordinated and timely actions based on real time information and preventive risk management are important prerequisites for efficient control of Varroa destructor. After a successful pilot project, the Austrian Beekeepers Federation (BÖ) together with other stakeholders (AGES, LKÖ) has launched a freely accessible, web based data sharing platform (www.bienengesundheit.at), which collects and analyses mite load data assessed by beekeepers enrolled in a sentinel apiary program.
A predictive algorithm extrapolates mite loads in individual colonies assessed with a standardised method (natural mite fall) at different times of the year. Based on these predictions the system performs risk assessments at a regional level. If mite loads are expected to exceed a predefined threshold, the system triggers an alert, which is subsequently published on different communications channels (social media, email, website). Predictions can be customized by the user to test the effect of treatments and brood rearing activity on mite populations. Based on weather forecast data, the system issues region specific recommendations for an efficient application of veterinary drugs whose efficacy is temperature dependent. The database includes >5200 samples from >780 colonies tagged with their geographic position and >70 beekeepers from 6 years. Currently, an API is being developed that will allow beekeepers using hive management software to transmit

mite counts to the database. This function is expected to further increase data flow to the platform.
In 2019, a newly developed diagnostic system (BeeVS®) using high resolution cameras for data acquisition and a cloud-based artificial intelligence for data analysis has been integrated in the sentinel apiary program. BeeVS® is able to detect and count Varroa mites on sticky boards in the field with high precision (94.3%) and sensitivity (94.7%), thereby assuring data traceability and data quality. The BeeVS® database includes >425.000 high resolution Varroa images.
The available mite load data will be used to validate and improve the prediction model and the alert function of the platform. In addition, the relationship between colony mite loads and weather will be analysed and results on the performance of BeeVS® will be presented.

[P.19.503] Detection of small hive beetle (Aethina tumida Murray) in naturally infested hives using DNA analysis of hive debris and scraps

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The small hive beetle (SHB), Aethina tumida, is a new threat of the beekeeping industry worldwide. The rapid detection of this pest at low levels of infestation is the key for eradication and early control, as visual inspection and the use of traps are time-consuming methods. The screening of hive debris collected from the bottom board is a promising detection method. We conducted conventional PCR analysis on hive debris collected from 60 infested and uninfested colonies and compared DNA extract to two SHB sets of primers. Results showed detection efficacy of 72.4% (IC 58.5-83.0%) for the long primers and 2.9% (IC0.4-18.1%) for the short primers. Detection efficacy of both primers was not correlated with the infestation level of the colony. Samples might be pooled by apiary or by beekeeper to assess the infestation status. This is an important improvement in efficacy compared to conventional methods of inspection that require time and trained inspectors.

[P.19.504] Standardized Human Assessments of Hive Health: Assessing the Healthy Colony Checklist and Hooper's 5 Questions as a Basis for a Standardized Bee Health Monitoring Tool

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With the many health challenges that honey bees are facing, beekeepers must check their colonies frequently to determine what management is needed to maintain colony health. Skilled observations and good recordkeeping are critical for tracking the efficacy of treatments and management. However, many beekeepers keep no records at all, and many of those that do keep records are both idiosyncratic and unsystematic. Very few go on to analyze which practices solve or prevent problems.
By establishing a standard for health assessment for honey bee colonies, we will provide beekeepers with a means of recording their observations and interventions that will a) lead to better colony care by individual beekeepers and b) enable the pooling and sharing of the inspection data on a large scale that will benefit not only beekeepers, but also the larger agricultural, scientific, business, and government communities.
This presentation focuses on building a standard health assessment that can be used by beekeepers everywhere. To do this we first propose a definition of a healthy colony and then compare existing checklists for assessing colony health including the Healthy Colony Checklist (HCC) and Hooper's 5 Questions (H5Q) being common ones among the several that can be found in the US and UK respectively.
The HCC and H5Q were developed independently; both were based on extensive experience and training, reviews of published works, and interactions with scientists and beekeepers. This study will provide an in-depth evaluation of hive inspection approaches to colony monitoring to ensure there are no significant gaps in coverage or relevance, but also the assumptions embedded in them, their intended purposes, and the methods for their proper use. The goal is to test and validate hive health assessment approaches to develop a standard that can be used to collect simple, understandable health assessment data that can be pooled, summarized, interpreted, and shared with beekeepers, researchers and policymakers to give them near real-time updates of honey bee colony health over time and in any region, country, or globally.

[P.19.505] Real-Time Diagnosis and Mapping of Emergent Honey Bee Health Issues via an AI-Powered App

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A newly developed smartphone app, using patented acoustic monitoring, has yielded a transformative way to improve honey bee colony health by letting honey bees communicate their health status directly to the beekeeper. In essence, the bees themselves become their own guru, indicating the status of colony health via the sounds they produce.
The Bee Health Guru smartphone app requires two steps: (1) Colony recordings are made by beekeepers using their smartphone microphone; and (2) Automatic diagnosis of a colony's condition is produced without beekeeper interpretation, via artificial intelligence (AI) software algorithms. The AI compares each beekeeper's recordings with specific archived audio recordings. The AI algorithms continue to learn, not only to diagnose specific situations in a beehive, but also to modify themselves, optimizing relevance for diagnosis and mapping

of emergent honey bee health issues.
The first step builds on a decade of referenced samples collected by research scientists and experienced beekeepers. It requests beekeepers download the application, inspect colonies and provide the app diagnosis along with their recordings. The second step allows observations to be easily submitted to our research scientists to supplement the diagnostics provided by the app. Based on a growing database of audio samples, paired with visual colony inspections, the Bee Health Guru smartphone application continually improves its own accuracy in diagnosing bee health factors, including varroa mites, foulbrood, nosema, queen status and other potential indications of overall colony condition.
In conclusion, the smartphone application automatically creates a copy of recordings and analyses, along with beekeeper observations, combining them into a comprehensive AI colony health diagnosis. It features real-time monitoring at the hive level and cloud-based data analytics to identify and map bee health issues. All electronic records are stored in a common destination (cloud) and have safeguards to protect data privacy, confidentiality, and security of beekeeper-reporters. The overall objective is to detect emergent colony health trends as they occur, monitor their geographic spread, and immediately share the results with citizen scientists and anyone else interested in honey bee health.

[P.19.506] A new electronic system to monitor the honeybee colonies

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The world of bees and honey are facing dramatic changes. Since the late 1990s, beekeepers around the world have observed mysterious and sudden disappearance of bees, and reported unusually high rates of decline in honeybee colonies. In the same time, the numerous studies show also a decline of other pollinators' populations. The main causes seldom incriminated are climate change, pesticide use, habitat loss, parasites, but looking at real issues leading to honeybees major reduction, the studies show that major problems are pesticides (52%), Asian hornet and other diseases and pests (i.e. Varroa) (32%), weather (16%), thief (10%). Taking into account that honeybees (*A. mellifera*) are a very good environmental biosensor, the development of an advanced electronic system that assess the development population status at any moment of the season is very important for environmental quality evaluation and for beekeeping management to prevent depopulation phenomenon. Therefore, the present studies aimed to develop a unique connected advanced solution for beehives to give direct information to beekeepers on pesticide contamination suspicion by evaluation the level of development of the bee colony population throughout the year, hornet attack detector, swarming signals and thief alarm. Two major features, as compared to market competitors, was developed on the new system: a device to count foragers bees therefore get many information about activity in and out the hive and an advanced sound analysis to detect hornet but also other signals connected with specific physiological status of honeybee colonies. The technical solutions and their efficiency will be analysed and discussed.

[P.19.507] Automated detection of swarm catch box occupation with the Internet of Things

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Regular monitoring of honey bee catch boxes is desirable to avoid an occupying swarm building large amounts of wild comb, necessitating a cut out. Catch boxes are placed around Australia's ports for biosecurity surveillance and remote inspection is advisable.
One method of connecting sensors to The Internet of Things is LoRa (LOng RAnge) radio networking, which utilises low power devices transmitting in the unlicensed spectrum transferring small "chirps" of data. LoRa devices are low cost and generally capable of communicating over ranges of 5-7 km depending on antenna configuration and line of sight. The LoRaWAN (LOng RAnge Wide Area Network) protocol has been extensively adopted and is supported by The Things Network (TTN), a system of open access public gateways. Network servers manage security and forwarding messages to application servers which process the data.
An Arduino-compatible microcontroller board (Rocketscream Mini Ultra Pro) with integral LoRa radio transceiver is equipped with ultrasonic and infrared distance-measuring sensors to monitor the internal space of the catch box. An occupying swarm triggers a change in distance to the reflecting surface. The device sends uplinks once per hour and data signals consisting of single byte messages are received by the TTN gateway. Messages are decoded by the bespoke TTN application which then forwards any positive results via text message or email, ensuring that detection of swarm lodging is reliable and timely. Battery life is estimated to be significantly greater than 1 year.
Further testing using an artificial swarm will be conducted to confirm detection for a live bee cluster. Extraneous insect or arachnid activity (such as spider webs) within the catch boxes may compromise measurements and necessitate periodic maintenance. Both IR and ultrasound sensors are being evaluated for robustness in field deployment.

[P.19.508] Software vs. Surveys: Comparing Approaches for Mapping Honey Bee Diseases

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As the world's population continues to rise, bees are more important than ever in guaranteeing a diverse food supply. Understanding where

bee diseases are, how they move, and how they impact colony health can help us manage the situation more intelligently.
Currently, we learn a majority of our knowledge about bee diseases from surveys, which give us great information for long term planning; however, often times there is significant damage already done by the time results are seen. Time is an extremely important factor when dealing with potential epidemics, therefore the immediate feedback available when using a software system provides the ability of quick reactions to information. Being able to make changes in time to prevent an outbreak and seeing if management choices work in real-time rather than later, are just a few of the benefits. Collecting bee data through a software system is a less intrusive process on the user and more granular information is received.
Utilizing data mining to determine what will be a problem before it is can only work within a large dataset of information. Rather than being surprised by a disease affecting bees in your providence, users will be able to see trouble coming and prepare in advance. Together we can build a future with enough data to see the impact of changes in treatment and policy.
In this study, we test survey metrics from traditional standardized survey sources against apiary management systems where beekeeper records are stored in a software system. We are testing data mining methods to see if they produce comparable information to the surveys. By comparing disease information from these different sources, we will not only be able to learn if using technology can give us a clear picture of the current state of bees around the world, but also the accuracy and validity of the data.

[P.19.509] How Honey Bee Colonies Can Benefit From E-Health - Introducing B-Keep®

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Today, beekeepers around the world are facing various challenges when managing their apiaries. From Varroosis and other pests and diseases to nutritional challenges and a changing climate, beekeepers must consider many variables to fulfil the needs of their colonies and to improve their chances of (winter-) survival. The many different tasks related to successfully managing honey bee colonies throughout the season require repeated on-site checkups and invasive monitoring of the hives.
The ongoing development of e-health tools and services in human and animal health, including honey bee health, can assist beekeepers in managing their colonies more efficiently. The short definition of e-health is that it refers to the digitalization of medicine. This includes the implementation of various digital tools and services, as well as digital data collection, analysis and archiving to improve prevention, diagnosis, monitoring and administration in healthcare.
Here, we would like to present B-Keep®, an innovative tool that supports beekeepers in detecting the needs of their colonies early and helps to limit the number of necessary on-site visits to the apiary. B-Keep® records temperature and humidity data of all connected hives and makes these data available to the beekeeper in a web application that can be accessed on a computer or mobile phone. Beekeepers are alerted when the collected data reach critical values in one or more of their colonies, allowing them to take preventive or reactive measures when necessary and making the management of their colonies more efficient.

[P.19.510] MiteCheck: A national strategy in the United States to reduce honey bee colony loss from varroa among small-scale beekeepers

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Many small-scale beekeepers in the United States lose colonies when their colonies become heavily infested with varroa. Some of these beekeepers are not aware of the need to manage mites, and others understand the need but do not feel confident in monitoring and managing the mites. In response to this, a group of programs and universities in the United States worked to develop a multi-level national training and support program to help small-scale beekeepers manage and monitor varroa. This program, MiteCheck, provides beekeepers tools to monitor varroa while simultaneously collecting data on national trends.
Many small-scale beekeepers report obstacles that keep them from regularly and efficiently monitoring for varroa in their hives: they do not realize that they have to monitor varroa, they do not have the equipment to monitor varroa, or they do not know how to/do not feel comfortable monitoring for varroa. MiteCheck specifically addresses each of these three obstacles by demonstrating the need and value of monitoring, providing access to kits that can be used to easily monitor, and providing training on how to monitor.
MiteCheck encourages small-scale beekeepers to report their varroa levels to an online platform at www.mitecheck.com. The MiteCheck online platform includes a map to demonstrate risk geographically and over time. As beekeepers report their mite counts, they can visualize the varroa risk in their region. The map acts as an early warning system for beekeepers when high mite levels are reported in their county, encouraging them to monitor their own colony mite levels.
MiteCheck has a mobile app, “mite-check,” available for Android and iOS phones. The app provides tutorials on how to test for varroa, a powdered sugar roll shake trainer, and an option to report mite levels and management strategies to the MiteCheck online platform. This app provides step-by-step instruction for beekeepers to help them effectively monitor mites.
Beekeepers may be interested in MiteCheck if they want to learn about methods for monitoring varroa. MiteCheck is also relevant for beekeeping instructors and leaders who want to provide resources and support to small-scale beekeepers to monitor and manage varroa.

[P.19.511] Towards an Electronic Nose for American Foulbrood

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American foulbrood (AFB), caused by *Paenibacillus larvae*, is the most economically and biologically devastating bacterial disease of honey bees. AFB is lethal to honey bee larvae, reducing them to a foul-smelling, glue-like mass, and subsequently causing colonies to die out. Early detection of AFB is crucial to prevent outbreaks from spreading to nearby beehives. However, current AFB field diagnostics rely on beekeepers first identifying symptoms, which may be as discreet as a single symptomatic larva in an apiary with hundreds of hives. Furthermore, the process of opening and inspecting hives can spread disease via equipment and is incredibly time-consuming, particularly for commercial beekeepers. Consequently, the apicultural industry requires rapid and non-invasive diagnostics for AFB. In recent decades, odour sensor systems ("electronic noses") have been used in the non-invasive detection of bacterial outbreaks in horticulture, food, and clinical settings. We are working to develop an electronic nose that can detect early infections of American foulbrood from volatile organic compounds in the hive air. This technology will be a valuable tool for the global beekeeping industry, providing a biosecurity device for screening hives for disease and allowing beekeepers to manage and respond to disease more effectively.

[P.19.512] Efficiency of Varroa monitoring methods - the benefits of standardized monitoring devices

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In the global debate about Varroa (Varroa destructor) management, treatments are often the main focus, but a well-organized monitoring routine lays the foundation for a successful treatment strategy. In a field study (2017) conducted in the Czech Republic, we compared the efficiency of four established Varroa monitoring methods: the two standardized, industrial methods "Varroa EasyCheck®" (Véto-pharma) and "Varroa Tester®" (Swienty) and the two home-made methods "Powdered Sugar Dusting" and "Alcohol Wash".

Ten beekeepers from all over the Czech Republic tested three out of the four monitoring methods on their own honey bee (*Apis mellifera*) colonies. Thus, three worker bee samples were taken from each colony on the same day between July and the end of August 2017. The efficiency of the monitoring methods was established by performing a second alcohol wash right after the first monitoring method had been applied on the same worker bee sample.

The mite infestation level of the colonies did not affect the efficiency of the four monitoring methods. The Varroa EasyCheck® and Varroa Tester® efficiencies were not affected by the factor "beekeeper" (the person who was performing the monitoring). The home-made Sugar Dusting method was significantly affected by the factor "beekeeper". The Varroa EasyCheck® and Varroa Tester® were the most efficient out of the four tested monitoring methods (82.3% and 83.9%, respectively), whereas the Sugar Dusting (64.9%) and the home-made Alcohol Wash (65.2%) methods demonstrated a significantly lower efficiency.

Our results give a clear indication of the benefits of standardized Varroa monitoring methods in comparison with home-made solutions, not only with regard to efficiency, but also in terms of reliability of results and usability by the beekeeper.

[P.19.513] Detecting colonies of Peanibacillus larvae larvae on MYPGP medium by semiconductor gas sensors

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The experiment was carried out in the laboratory conditions in two chambers with a wooden or a polystyrene interior. They were connected to a multi-sensor, air quality detector. The tool was based on the semiconductor FIGARO sensors: TGS623, TGS823, TGS826, TGS832, TGS2600, TGS2602, TGS2603.

The aim of the experiment was to determine to what extent and at what stage of bacterial growth semiconductor gas sensors are able to detect colonies of Peanibacillus larvae larvae.

Gas samples from clean MYPGP medium and from medium with P. l. larvae culture (ATCC strain 9545, ERIC I) were tested after 1 to 10 days of incubation at 26°C. As a control, air from an empty test chamber was used. The exposure phase of the test sample lasted 10 min, the tests were carried out at 21 ° C. The sensor response was analyzed in 270s.

The gas samples taken from MYPGP medium with P. l. larvae culture, after the first day of incubation, gave an average higher sensor reading (from 0.508 V for TGS823 to 2.324 V for TGS2602) than gas samples taken from a clean base (from 0.364 V for the TGS823 sensor to 2.2 V for the TGS2602 sensor), but lower than air from the MYPGP medium with P. l. larvae culture after 2 days of incubation. Similar relations were found in the chamber with the polystyrene interior.

It was shown that the multi-sensor air quality detector used in the study detects the presence of colonies produced by P. l. larvae bacteria on the MYPGP medium. However, after the second day of the plating, the sensors do not differentiate the colony's age. The test results provide the grounds for the use of semiconductor gas sensors in the detection of dangerous diseases of sealed brood.

[P.19.514] Use of digital technologies to strengthening of beekeeping in Santa Catarina state, Brazil

E. Blainski, D. Dortzbach, E. Salum Pereira Pinto
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Brazil has an international highlight for the production of honey with different characteristics. In all regions of the country, beekeeping has been important as a complementary rural activity, especially in cases of family farming. Despite the great importance of bees for the generation of wealth and for the realization of ecosystem services (pollinating agents), the activity has systematically suffered losses attributed mainly to use of pesticides in the management of crops, to degradation of forest remnants, to burning of forests, to expansion of agricultural and livestock areas and to climate changes. In this work, the aim was to develop tools for the monitoring, storage and processing of data related to bee behavior and for the dissemination of technical information able to assisting beekeepers in decision-making looking for sustainability of the activity in the Santa Catarina state, Brazil. For this, automated data collection platforms (DCP) were developed with data transmission system that enabled the real-time monitoring of the behavior of beehives located in six agroecological regions of Santa Catarina state. With the DCP's it was possible to monitoring the weight gain of the beehives, the temperature and relative humidity inside the beehives, the temperature and relative humidity of the air and the rain. In addition, it was make the monitoring of food supply inside of the area of bees activity (3 km). The data analysis was made to defined the seasonality of honey production in the different agro ecological regions. For the information diffusion, a web system was developed that allows direct contact between beekeepers, researchers, technicians and other actors involved in the beekeeping chain. The results of the environmental monitoring allowed to identify differentiated patterns in honey production, influenced by the land use and climatic variability of Santa Catarina state. The production of honey showed a correlation with the available food supply in each of the six agroecological regions monitored. The online system to information dissemination has proven to be effective in assisting the beekeeper for the management of beehives and an important technical assistance tool. The system can be accessed in: http://ciram.epagri.sc.gov.br/apicultura/.

[P.19.515] Application of geoprocessing to delimit potential areas for organic honey production in the Santa Catarina State, Brazil

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Santa Catarina state is located in the southern region of Brazil and has great prominence due to the large number of beekeepers. According to data from the Federation of Beekeepers Associations of Santa Catarina (FAASC), the state has about 6,000 beekeepers who make the state the largest national honey producer per km², the largest exporter of the product and, moreover, Santa Catarina is a world leader in diversity and quality of the honeys produced. Besides of honey production, the migratory beekeeping is an important activity where the main product is the pollination service provided by the bees. The apple crop is the main demander of these services, annually the sector invests around R $ 3.2 million in the rental of 45 thousand hives that are responsible for the pollination of approximately 90% of the orchards of Santa Catarina. However, even with bee importance for different sectors of the economy, cases of mortality and contamination of honey by pesticide residues have become common in Brazil and in the world. In Santa Catarina this problem has harmeding the beekeeping sector and imparing the production of differentiated honeys like as organic. In this work, the aim was to develop a tool that allows the georeferencing of apiaries and the correlation of these data with images of land use. This results was used to delimit potencial areas to the organic beekeeping activity and to identify areas of conflicts of interest. The tool was initially used in the northern region of Santa Catarina. It was possible to mapping the regions with conflicts of use between beekeeping and agriculture. In addition, it was possible to identify and mapping areas suitable for organic beekeeping. The technology allows monthly updates, so it was possible to monitoring the landscape changes and thus adopt protective measures. The results of this research can be used to establish public policies aimed at the preservation of bees.

[P.19.516] Fluctuating asymmetry as environmental bioindicator: evaluation of the asymmetry of the wings of *Melipona seminigra pernigra* through geometric morphometrics

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Melipona seminigra pernigra is one of the most abundant stingless bees in the East Amazon and is economically important for honey production and pollination. In natural environments, the bees develop and live under disturbances, which can cause certain instability in the floating asymmetry of wings venation. We, collected worker bees from 10 colonies of M. seminigra, installed in different locations: pristine forest, forest near human facilities (buildings and road) and a forest bordering an open pit iron mine. All locations were in the Carajás National Forest, Pará, Brazil, sampling occurred between March and May 2018. The anterior right and left wings were removed, mounted between microscope slides and photographed. We digitized 12 anatomic landmarks at the intersections of the veins using the tpsDig program. Through the Procrustes ANOVA, and ANOVA two-factor we obtained the symmetrical and asymmetric components of the variables of shape and size, followed by principal component analysis. The Procrustres ANOVA for the coordinates of shape and ANOVA two-factors for the size of the centroid indicated that the workers of M. seminigra presented fluctuating asymmetry of shape and size

statistically significant in the populations of the three localities sampled. The presence of directional shape asymmetry was also observed. However, we found no directional size asymmetry in the individuals from the three situations tested. The patterns of individual variation and fluctuating asymmetry were exhibited from the principal component analysis. The first two main components (PCs) were required to explain about 50% of the shape variation in populations analyzed, except for the meliponary population of the mine area, where the percentage of 58% of the total variation was explained by the first PC. Based on the percentage of total variation displayed by the PCs, populations of M. seminigra showed similar fluctuating asymmetry values in the three landscapes studied.

POLLINATION AND BEE FLORA

09 SEPTEMBER 2019

POSTER SESSION 04 08:30-18:00
POLLINATION AND BEE FLORA I POSTER AREA

[P.04.95] Neotropical Bees: integrating scientific knowledge through an information system

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Brazil has made great progress in the organization and open sharing of data on the taxonomy and occurrence of Neotropical bees. In order to provide data on bee species in an integrated and fast manner, the Brazilian Bee Studies Association (A.B.E.L.H.A., acronym in Portuguese), in partnership with the Reference Center for Environmental Information (CRIA, acronym in Portuguese), launched in 2016 the Scientific Information System on Neotropical Bees. The system uses Moure's Bee Catalog as its primary source of scientific, common, and indigenous names, for searching and retrieving data from different public sources of information in Brazil and abroad. The current version of Moure's Bee Catalog (2013) recognizes more than five thousand species of Neotropical bees and shall be updated by the end of 2020. One of the sources of data shared through the Information System is speciesLink, a network that serves species occurrence data provided by bee collections from Brazil and abroad. Currently, speciesLink integrates more than 290 thousand online records of 1,479 species of bees, of which 1,256 occur in Brazil. In addition to occurrence data, speciesLink also shares about 7 thousand images of vouchers and live material. The Information System also presents scientific images from Flickr and data on bee-plant interactions for more than 900 species of bees and two thousand species of plants. It also integrates data served by bibliographic databases such as the Biodiversity Heritage Library, Bioline International, Directory of Open Access Journals, and the Brazilian Portal of Scientific Publications in Open Access - oasisbr. When carrying out a search by species, users access data from all these different sources on a single page and can always access the original source for more information. By integrating data from various sources, the Information System provides a fast and more efficient way to access information on bees, improving the comprehension on native bees, where they are found, which plants they visit, and which studies exist in Brazil. The Information System is accessible to all interested parties at http://abelha.cria.org.br/.

[P.04.96] *Apis florea* Invasion in Sudan: Present Status and Future Implication

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Abstract: The exotic dwarf or a little honeybee, *Apis florea*, is originally prevalent in tropical and subtropical of Southeast Asia regions. The widespread distribution of this bee is limited to warm climates where it performs excellent. Over the past decades A. florea has been accidentally introduced by human activities to Africa subcontinent where the species established sustainable and expanding vital populations very well. In 1985 A. florea has been detected in a garden in a suburb and forest of Khartoum in Sudan. Invasion and rapid expansion of A. florea into Sudan region have shown significant and effective colonizing ability, as well and its dramatic contribution on agri-ecosystem. However, the species is well adapted to new hot arid conditions of Sudan without being affected by the competition from others wild honeybee species. It is likely that A. florea in Sudan might have undergone certain behavioural changes or degeneration in its characters during ecological adaptation to different climatic conditions and excessive inbreeding after its introduction into Sudan. Therefore, this review is trying to study the present status and detailed research of A. florea and to understand its impact in the new ecosystem and potentially possible economic advantages.

[P.04.97] Pollinators Resources Vis-a-Vis Sustainable Crop Production in Developing Countries

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Insect pollinators are indispensible in global agriculture for their valuable ecosystem services. The estimated economic value of the insect pollination services is about € 153 billion. Three quarters of the world's cultivated crops are pollinated by bees as most effective and reliable pollinators.
In biodiversity rich country like India, insect pollination services have not received much attention in crop production process. However, recent reports indicating decline in the abundance of wild pollinator population owing to modern agricultural practices is of much concern for Indian agriculture. Potential benefits of and qualitative yield parameters of many cropping systems have been well documented In apple crop alone, the managed bee pollination has increased the fruit set by 9.09 per cent with an estimated economic benefit of Rs.22000/- for individual farmer per acre of apple orchard. In case of Apple, pomegranate and onion the reported gains in the crop production due to managed bee pollination was about 34.89 and 17.21 per cent with corresponding economic gains of 42.02 and 18.51 per cent, respectively. Annual demand of bee colonies for the pollination services was estimated about 200 million colonies considering the area

under cultivation of cross pollinated crops. The concept of renting of bee colonies is in nascent stage and it will further rise as most of the crops have reached their yield plateau despite the good agricultural practices. Policy directives for incorporation managed bee pollination as a component of plant protection for sustaining and enhancing the crop productivity is the need of the hour.

[P.04.98] Bees or no bees,this is the question: global educational challenge in the direction of saving the bees and environment conservation

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The Brazilian NGO Bee Or Not to Be concerned with the increasing mortality of bees in the world and, especially, in Brazil, decided to call the attention of researchers, authorities and entities beekeeping to the need for urgent development of environmental education programs in schools. Without this initiative, we do not see the possibility to protect them and conserve the environment. This action should be directed to children, in whom we hope for our future. In this sense, the NGO of Brazil, in 2018, launched an educational program entitled Environmental Education Project No Bees, No Food, based on the didactic work of the same name No Bees, No Food: Activity Book for Environmental Education, written by Rosane M. G. Peruchi and Lionel S. Gonçalves (2nd edition, 2017). The project was implemented during three months in the city of Ribeirao Preto-SP, with support from the Secretariat of Education and the Secretariat of the Environment, under the coordination of Dr. Rosane M. G. Peruchi and Dr. Maria Juliana F. Caliman, in the municipal network. 21 municipal public schools have participated in the Project, reaching 1,821 students all in the 3rd grade of Elementary School, children about 8- 9 years. The program was divided into 5 modules: What are bees like (morphology and biodiversity); Where do bees live (bees depend on flowers); What is life like in a beehive (basic biology of bees); Why are bees so important (the concept of pollination and notions of sustainability); and How can we protect the bees. The didactic approach highlights the service of pollination, that contributes to the feeding of animals, of man and for the conservation through the reproduction of plants. The project aims to gather arguments on how to protect them, including keep our environment (water, soil, air) healthy and sustainable for the future. The reaction of the children was amazing and we believe that environmental education projects through the studies of bees can be a great facilitator for global educational challenge in saving the bees, environment conservation and improving the quality of life on our planet.

[P.04.99] Diversity and abundance of wild bees in Mexico's Nevado de Colima, national park

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Bees, the main pollinators of flowering plants, are classified into seven families. In Mexico, there is a wide diversity of them, since there are 1840 species within six families. The objective of this study was to determine the diversity and abundance of wild bees in Mexico's Nevado de Colima National Park. The climate at the park varies from temperate to cold, with a yearly average temperature of 10° C. Bees were collected from flowering plants during four seasons of a year, at three different altitudes, 1900, 2600 and 3300 meters above sea level (masl). We obtained 1311 bee specimens belonging to five families. In those specimens, we identified 35 genera and 59 species, of which, eight are social (13.6%) and 51 are solitary (86.4%). The families with the highest specific richness were Apidae (30) and Halictidae (7). Two endemic species of stingless bees (*Melipona colimana* and Plebeia manantlensis) and three kleptoparasitic species (Coelioxys spp., Nomada spp., Sphecodes spp.) were found. At 1900 masl, there was a greater diversity of bees (51 spp.) as well as greater abundance (66% of the specimens), followed by 2600 and 3300 masl, where there were 14 and 11 spp., and 11 and 23% of the specimens collected, respectively. The bees that visited more species of flowering plants were *Apis mellifera* (51 plants), Bombus ephipiatus (27), Ceratina capitosa (20) and five species of the genus Lasioglossum (19). The highest abundance of bees was observed during fall and part of winter, the time of the year when more plant species were blooming. The diversity of bee families found at Mexico's Nevado de Colima National Park is high, although it is relatively low at the species level in comparison with tropical and arid zones. Edemism of some species was evident. The implications of these findings will be discussed

[P.04.100] Sexual Maturity Time of Reproductive Organ Development and Mating in the Korean Native Bumblebee, Bombus ignitus

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To increase the mating rate of *Bombus ignitus* used as insect pollinator, we investigated the sexual maturity time of B. ignitus. In investigating ovary development such as the number of eggs per ovariole and spermatheca size, the time of sexual maturity of queen was 10 days after eclosion. In case of male, the number of sperm was 246 thousand at immediately after eclosion, and was highest as 480 thousand at 9 days, and tended to show a dramatic decline at 35 days (87 thousand). The more mating time, the less the number of sperm. In consideration of number of sperms, the time of sexual maturity of male was 3-15 days after eclosion. In the sexual maturity

time of queen in mating, the queen was not mated at immediately after eclosion, and showed a decrease in 20 days. In terms of the rate of mating and oviposition, the favorable time for mating of queen was 9-20 days. On the other hand, the male showed 3.3% of mating at immediately after ecolosion, showed the highest of 43.3% at 6 days after eclosion, and tended to decrease in 25 days. The sexual maturity time for mating of male was 6-20 days. In summary, our results indicate that sexual maturity time of B. ignitus in reproductive organs and mating is most favorable in 9-12 days after eclosion for queen and 6-9 days for male.

[P.04.101] Morphometry of Ceiba pubiflora (A.St.-Hil.) K.Schum flowers

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Morphometric analyzes of flowers are important in identifying potential floral visitors and potential pollinators, as well as collaborating with information related to reproduction, genetic maintenance, preservation and perpetuation of angiosperms. In this context, the aim of this study was to evaluate the morphometry of Ceiba pubiflora (A.St.-Hil.) K.Schum flowers. This tree species is native to the Brazilian South and Southeastern region and used in urban afforestation, in large places, due to its large size.The study was conducted from April to May 2018 on the campus of the Maringá State University. Morphometry analysis was performed on two flowers in anthesis per tree, totaling 20 flowers in ten trees, with a pachymeter. From each flower was measured total length (base of the chalice to the apex of the corolla); chalice length; corolla diameter; number, length and width of petals; stamens and the diameter of the stigma. Means of each analyzed parameter and their respective standard deviations were obtained with the statistical software R (R Development Core Team). The data revealed pentamerous flowers, with mean length of 11.09 ± 3.17 cm; chalice of 3.05 ± 0.26 cm in length; corolla diameter of 15.33 ± 1.38 cm; length and width of the petals of 9.44 ± 0.96 cm and 4.51 ± 0.45 cm, respectively. For reproductive structures, the stigma diameter of 0.19 ± 0.02 cm; length of the stamens of 6.32 ± 0.37 cm and length and width of the anthers of 0.64 ± 0.04 cm and 0.44 ± 0.03, respectively. The nectary stays at the base of the ovary and produces large amounts of nectar per flower. Pollen grains, in abundance, are also available to flower visitors. Due to the large size of the flowers, they are likely to be visited by a variety of insects, birds and small mammals, but having as their likely potential pollinator large bee species, such as Bombus spp. and Xylocopa spp., butterflies, moth, hummingbirds and/or owls.

POLLINATION AND BEE FLORA

10 SEPTEMBER 2019

POSTER SESSION 10 08:30-18:00

POLLINATION AND BEE FLORA II POSTER AREA

[P.10.199] Honeybee pollination enhances the sensory quality of melon fruit (Cucurbitaceae: Cucumis melo L.)

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Fruit size, sugar contents and volatile aromas are key focuses in fruit breeding. The present study was conducted to compare fruit development, fruit sugar content and volatile aroma profiles between honeybee pollinated and forchlorfenuron treated melon fruit (Cucumis melo L.). Our result demonstrated that forchlorfenuron-treated melon fruits exhibited significantly increased individual fruit weight, and fruit size with cracked fruit surfaces. In contrast, honeybee-pollinated fruits were characterized by relatively medium size, uniform fruit surface networking, a higher seeds weight, and higher total sugar content. Furthermore, honeybee-pollinated melon fruits were characterized by higher ester volatile content, which are important representatives of the aroma profile. However, forchlorfenuron-treated melon fruit were characterized by relatively higher aldehydes and ketones volatile reportedly distinguished in seedless and non-mature melon fruit. We conclude that the use of the honeybee in greenhouse melon pollination may be an effective approach for obtaining a desirable appearance and sensory quality of melon fruit.

[P.10.200] Managed honeybees (*Apis mellifera L.*) increase onion (Alliun cepa) seed yield and quality

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Nearly 75% of the world's flowering plants are dependent on insects for pollination, with honeybees being well known for their importance for several crops. The effect of managed honeybee pollination on onion seed yield and quality was investigated through pollinator exclusion and pollinator surveys on onion field plots at Mekelle Agricultural Research Center experimental farm. The treatments were: plots accessible to all flower visitors (CTL); plots not accessible to any insects – the plots were covered with an insect proof mesh cage before the ray florets started opening (NI); plots accessible only to honeybees – the plots were covered with an insect proof mesh cage and a honeybee colony with four frames was placed inside the cage during the flowering peak (HB). Insect proof mesh cages (5m x 3m and 2.5m high) were made of wood covered with 20% shade cloth. All insects were removed from all the cages before blooming, to exclude unwanted pollinators. Honeybee colonies used in this experiment received supplementary feeding (dissolved sugar) and water before and after they were placed in the cages. The results revealed that onion seed yield was increased by 41.2%, the mass of 1000 seeds by 25% and germination percentage by 68% in the open pollination especially with honeybees. Open pollination treatments especially with honeybees increased onion seed quantity and quality.

[P.10.201] Foraging behavior of honeybee (*Apis mellifera*) on flowers of Guzzotia abyssinica and its impact on seed yield and quality, Mekelle, Ethiopia

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The study was conducted at Mekelle Agricultural Research Center with the objective of determining the flight intensity and foraging behavior of local honeybees in relation to climatic factors. The study was also designed to evaluate the effect of honeybees' pollination on yield and quality of G. abyssinica seeds. Evaluating of honeybee's pollination was conducted in a complete randomized block design with three treatments and four replications. The treatments were caged with honeybee, caged without honeybee and open pollinated. The foraging behavior of bees was conducted in the open plots. The flight intensity of honeybees was determined by placing four honeybee colonies. The obtained data related seed yield and quality parameters were statistically analyzed using one-way ANOVA. Data related flight intensity and foraging behaviors of honeybees as a function of time of the day were analyzed using GLM repeated measurement analysis procedure.

Significantly the highest seed yield per plot (1.2m2) was found in plants caged with honeybees (1669gm), while plants excluded from insects had the lowest yield (960gm). The highest numbers of honeybees were recorded at 8:30-9:30AM (34), while the least number of bees were recorded at 16:30-17:30PM (1.5). The number of bees that collected nectar had a positive association with air temperature ($r=+0.67$; $P=0.01$), while negative relationship with relative humidity ($r=-0.59$; $P=0.001$). However, the number of bees that collected pollen had a positive correlation with relative humidity ($r=+0.62$; $P=0.001$), while negative association with air temperature ($r=-0.72$; $P=0.001$). The peak in the number of bees that exited (245) and returned (218) was recorded at 9:30AM, whereas the least number of bees that exited (66) and returned (54) was recorded at 7:00AM. The number of bees that exited and returned were positively correlated with air temperature and sunshine hours, and a negatively correlated with relative humidity and cloudiness. Therefore, this pollination

technology to G.abyssinica seed producers is highly recommended and honeybee pollination should be considered as one of the inputs for G.abyssinica seed production. Honeybee and other insect pollinators can therefore be saved from pesticide hazards by not applying at times when flight and foraging activity of bees is highest.

[P.10.202] Impact of beekeeping with Indigenous bees (*Apis cerana* & *Trigona*) on Mango and Cashew production in India

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Since bees play a crucial role in increasing agricultural productivity, Under The Mango Tree (UTMT) Society launched the Bees for Poverty Reduction (BPR) programme in 2009 which targets rural poverty through beekeeping with *Indigenous bees*. The income earned from enhanced productivity and sale of agriculture commodities adds a crucial source of income for smallholders. This paper will assess the impact of beekeeping with *Indigenous bees* on pollination of Mango and Cashew in two locations – Valsad (Gujarat) and Palghar (Maharashtra) – in western India. Data collected (in 2018-19) from a set of 50 farmers, those who have beeboxes of *Apis cerana* and *Trigona*, will be compared to another set of 50 farmers who do not have beeboxes. The control group farmers will be selected from the same geographical area, with similar agro-climatic conditions, who followed the same agricultural practices, such as irrigation, usage of fertilizers, and pesticides. It will be ensured that farmers who have Mango and Cashew orchards, and have been already trained in beekeeping by UTMT Society, have filled beeboxes (*Apis cerana* or *Trigona*) in their farms or orchards during the flowering season. The paper will present the findings of three different types of comparison: 1) Pre and post beebox crop output of Mango and Cashew; 2) Fruits pollinated by *Apis cerana* and *Trigona* species of bees; and 3) Pollination impact in two different geographies - Valsad (Gujarat) and Palghar (Maharashtra).
This kind of pollination impact in farmer field situations with *Indigenous bees* is being studied for the first time in this part of India, and the research has been supported by Bees Abroad, UK.

[P.10.203] Foraging behaviour of *Apis mellifera L.* and *Scaptotrigona bipunctata* in the Dombeya wallichii

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The visitation activity and behavior of pollinators in flowers is influenced by several factors for example temperature and relative humidity and plant characteristics such as the rewards offered by the flower, which are rich in energy, minerals, proteins and vitamins and contribute to the nutrition and health of bees. Due to the lack of information on the interaction between the flower of Dombeya wallichii and its flower visitors, the behavior of the species *Apis mellifera* and *Scaptotrigona bipunctata* in the flower of Dombeya wallichii during the period of 8 am to 17pm was evaluated during three days, observing a bee for a minute to total ten bees per hour. For each visit, variables were recorded: type of floral resource collected (nectar or pollen), number of flowers visited and length of stay of the bee in the flower. In total, 291 honey bees africanized were observed, of which 74.3% collected nectar, 4.7% pollen and 21% both resources.It was observed that at 1:00 half of the bees collected nectar and the other half nectar and pollen while at 16 hours, all the bees were collecting only nectar. This species visits on average 4.2 flowers per minute with a duration of 9.2 seconds per visit. In relation to S. bipunctata, 239 forager bees were analyzed, distributed in percentages of 69.3%, 17% and 13.7% for collection of nectar, pollen and both resources, respectively. The species visited 1.8 flowers per minute with an average duration of 33.8 seconds per visit. From the floral visitors studied, *A. mellifera* was noted for its effectiveness because during its visit it touched both the anthers and the stigma of the flower and visited a greater number of flowers per minute, but S. bipunctata was characterized by being larger time visiting the flower. Regardless of the species and time evaluated, a preference was observed for the collection of nectar or nectar and pollen.

[P.10.204] The impact of lowbush blueberry (Ericales: Ericaceae) and cranberry pollination on honey bee (Hymenoptera: Apidae) colony health status

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Commercial lowbush blueberry (Vaccinium angustifolium Ait.) and cranberry (Vaccinium macrocarpon Ait.) crops require the honey bee (*Apis mellifera L.*) for pollination. Unfortunately, beekeepers are observing negative impacts of pollination services on honey bee colonies. In this study, we investigated three beekeeping pollination management strategies (MS) and measured their impact on honey bee colony health and development. Experimental groups were: A) control farmland honey producing MS; B) Blueberry pollination MS; C) Cranberry pollination MS and D) Double pollination MS, blueberry followed by cranberry. Our goals were to 1) compare floral abundance and attractiveness to honey bees between apiaries using a Geographic Information System, and 2) compare honey bee colony health status and development between MS during a complete beekeeping season. Our results show significantly lower floral abundance and honey bee attractiveness during cranberry pollination compared to the other environments. The double pollination MS significantly reduced colony weight gain, pollen harvest and brood population, and induced a significantly higher winter mortality rate. We also measured significantly higher levels of Black queen cell virus, Sac brood virus and Nosema ceranae in the double pollination MS.

[P.10.205] Production of pods in soybean (Glycine max) with and without africanized honeybee colonies

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In pollination services, bees are encouraged to collect pollen during the flowering of the crop of economic interest. Regarding soybean (Glycine max), there are very discrepant references regarding the contribution of pollinators, especially *Apis mellifera*, varying from the absence of contribution to increases of more than 30% in the yield of plots with adequate honeybee visitation. Based on this, the present study was carried out to evaluate the number of pods per plant and the weight of pods with one, two, three and four seeds in different cultivars. The experiment was conducted at the Experimental Station of Embrapa Soja in Londrina city – State of Paraná, South of Brazil, and the cultivars used were BRS 1001 IPRO and BRS 284. Pollinating cages were installed in covered areas, 4 m wide, 6 m long and 2 m high at the top, covering an area of 24 m². Approximately one week before the expected start of flowering, the cages were assembled for developing the experiment and after this time disassembled, allowing the full vegetative development of the plants. We observed that the covered areaswith Africanized honeybee colony produced approximately 20% more pods per plant than the coveredareas without honeybee colony. The main difference in production between covered area with honeybees and the covered area without honeybees is mainly due to the higher weight of the seeds in pods with three and four grain found in the coveredareas with honeybee colony. Therefore, we affirm that even the soybean - Glycine max being an autogamous plant, in the treatments with the Africanized honeybee colony- *Apis mellifera*,it was obtained higher results than the number of pods per plant and seed weight in the BRS 1001 IPRO and BRS 284 cultivars, confirming that honeybee pollination has influence on soybean crop productivity. Because of this, this plant has a wide range of the honeybee pollination effect that depends on the cultivar used for the experiment and the results cannot be generalized for all cultivar of soybean, but specifically for one of that assay.

[P.10.206] Soybean and bees in Brazil

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Soybean is progressively becoming a forage option for both wild and domestic (*Apis mellifera*) bees in Brazil. The soybean area in the country surpassed 36 million hectares in the 2018/19 season, so the cropped area is getting closer to the apiaries, as well as bee keepers are placing their hives next to soybean fields, during its blooming. To understand the intensity and preferred time of the visitation of bees on soybean, assessments were made on seven different Brazilian locations: Passo Fundo-RS (-28.2260; -52.3267), Cerro Largo-RS (-28.1677; -54.7201), Londrina-PR (-23.2017; -51.1879), Dourados-MS (-22.2733; -54.8241), Campo Novo do Parecis-MT (-12.5748; -58.1026), Sorriso-MT (-12.5361; -55.5900) and Luis Eduardo Magalhães-BA (-12.0093; -45.7179). Results indicated that more than 40 bee species visited soybean fields, and the prevalence of the species depended on the vicinity to apiaries and the dimension of native forests on the landscape. Domestic bee were more frequent on the majority of the locations, while non-Apis species were predominant on the two MT locations. The visitation of bees largely (ca. 90%) occurred during the morning. In general, higher frequency of *Apis mellifera* visitation on soybean was observed from 9 to 11 a.m., while wild species showed no preference, without a clear population peak. Studies to establish the intensity of bee visitation, according to the distance from the border of the field, demonstrating that over 80% of the population concentrated up to 200m from the border. In order to avoid any non-targeted effect of phytosanitary measures used to control soybean pests on the foraging bees, soybean growers are oriented to avoid insecticide application during soybean blooming. If an application is technically recommended then it should be performed late in the afternoon or at night, also avoiding sprayings close to the border of the field. The growers are also instructed to communicate neighboring beekeepers at least 24h before any pesticide application, in order to to allow them to close their hives the night before the application.

[P.10.207] Floral sources explored by honeybees (*Apis mellifera*) and stingless bees (*Scaptotrigona depilis*) in coffee (Coffea arabica) crops intelligent and assisted pollinated by AgroBee®

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The pollination value was estimated at about US$ 12 billion in Brazil in 2018, but the potential of assisted and intelligent pollination is still poorly explored in the country. About 85 agricultural crops are benefited directly or indirectly by bee pollination, and the coffee (Coffeea arabica and C. canephora, Rubiaceae) is one of these crops. The Brazil is the world's leading coffee producer and exporter: in 2018, almost 60 million of sacks of 60 kg were produced. Although it does not require pollination by animals, studies have already shown that the presence of bees pollinating the flowers increases the amount of fruit produced as well as increasing the fruit size by up to 30%. This study proposed to investigate the floral sources explored by two species of bees with management potential, *Apis mellifera* (Apini) and *Scaptotrigona depilis* (Meliponini), in association with coffee (C. arabica) crops. Four rural properties of southeastern Brazil were studied, and at least 10 foraging workers of each species were randomly sampled during the coffee's blooming in September 2018 in each property. The pollen of the body and corbicula of these workers were submitted to acetolysis process and the pollen types were

identified using comparative method with the reference material. Sixteen pollen types besides C. arabica of 11 families were identified, being Fabaceae and Myrtaceae the most representatives. The pollen of coffee was 'very frequent' (> 45%) or 'frequent' (16-45%) in the diet of A. melifera and S. depilis in three and two studied proprieties, respectively. As the coffee honey is also an appreciated product, the management of these bees associated with this crop can be an interesting activity also for the beekeeper. Other important food sources were species of Eucalyptus (Myrtaceae) for both bee species, and type Bidens (Asteraceae) and type Gliricidia (Fabaceae) for *A. mellifera* and S. depilis, respectively. Added to the great knowledge in the management of both species, this result indicate that these studied bee species have great potential in the use of assisted and intelligent pollination (AgroBee®) of coffee crops.

[P.10.208] Effectiveness of Nectar Contents on the Foraging Activity of *Apis mellifera* on Asian pear (Pyrus pyrifolia Nakai)

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The foraging preferences of *Apis mellifera* for flower depend on the content of sugar and amino acid in nectar. To determine the factor in nectar that attracts honeybee in field, we investigated the foraging rate of honeybee, fruit set, relationship between the foraging behavior of honeybee and contents of nectar in different Asian pear cultivars ('Niitaka' and 'Whangkeumbae'), which do not produce pollen. The foraging rate of honeybee on 'Whangkeumbae' was 2 times higher than that on 'Niitaka' and the fruit set of 'Whangkeumbae' was 1.5 times higher than that of 'Niitaka'. 'Whangkeumbae' exhibited 3.7 times higher nectar secretion than 'Niitaka'. The total sugar content in the nectar of 'Whangkeumbae' was 1.1–1.7 times higher than that in the nectar of 'Niitaka', and this difference was significant. Furthermore, the content of glucose, fructose, and sucrose in the nectar of 'Whangkeumbae' was higher than that in the nectar of 'Niitaka'. In particular, the content of sucrose in 'Whangkeumbae' was 1.3–3.2 times higher than that in 'Niitaka'; sucrose is known to be the most preferred sugar by honeybee. Among the 20 amino acids detected in both cultivars, the content of Nine amino acids was significantly different. The content of phenylalanine and glycine (which are preferred by honeybee) in 'Whangkeumbae' was 1.4–2.4 times higher than that in 'Niitaka'. In contrast, the content of asparagine (which is not preferred by honeybee) in 'Niitaka' was 1.1–2.4 times higher than that in 'Whangkeumbae'. Among the sugars in nectar, sucrose exhibited the highest relationship with the number of foraging bees, followed by glucose and fructose. Among the amino acids, phenylalanine presented the highest correlation with the number of foraging bees, followed by glycine and alanine. The results suggest that honeybees prefer 'Whangkeumbae' over 'Niitaka' because of the difference in the amino acid and sugar content in the nectar of pear cultivars.

[P.10.209] Using bees (Hymenoptera, Apidae) for coffee's (Coffea arabica) crop intelligent and assisted pollination (AgroBee®) in different regions of Brazil

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75% of the world's food comes from animal pollination, and in Brazil, about 85 agricultural crops are benefited by bee pollination. The incorrect application of pesticides, among other anthropogenic actions, resulted in a decrease in the bee population, with the need for the implementation of assisted pollination as a secondary resource to increase the productivity and quality of agricultural crops. The coffee crop (Coffea sp.) chosen because Brazil is one of the largest producers and exporters of the grain and although this crop does not depend of the bees for exist. Studies indicated an increase of up to 30% in productivity when these insects inserted in the crop. Offering the intelligent and assisted pollination service to farmers could increase their production without having to increase the area of planting this grain, a friendly environment way to increase the quantity of food, and this was the focus of this work (AgroBee® Plataform/App). Four selected properties, (A) São Sebastião Paraíso/MG, B1 and B2 - Uberlândia/MG, C - Ribeirão Corrente/SP (1 hect/locality). Within the permanent protection areas, 10 colonies of 2 bees species, *Apis mellifera* and *Scaptotrigona depilis* were inserted. They marked with colored ribbons, 30 coffee branches per distance (10m; 50m;100m) from the place where the bees were and 30 coffee branches were isolated with voil. There was a low production of fruits in the branches that covered in relation to those not covered. The branches marked at 10m showed uniformity in the highest fruiting in relation to the branches marked at 50m. At 100m, fruit and fruit properties were observed in the A and C properties. However, in the B1 and B2 properties, both small areas with a good forest around, the benefits of the additon of new colonies were not so detected. Therefore, the insertion of colonies of bee species that are efficient pollinators of the coffee helps to increase the production of the grain, when the colonies placed in places near the crop.

[P.10.212] Complementation of Stingless, Solidary and Honey bees, in both the Greenhouse And In Open Field Pollination For Sustainable Food And Nutrition Security In Kenya

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Stingless, solidary, as well as honey bees, are vital in social, economic and cultural development of most communities in the developing world, particularly in tropical regions. It oscillates around the past, present and future, expressed in conservation, biodiversity and sustainability. The art

of keeping bees encompasses inter-linked processes. For profitable beekeeping, technology and creativity shall be at the fore, right from honey production, processing, value addition and marketing, with employment prospects. It had been observed that stingless bees (Jaycox, 1979; Sellers, 2007), alongside honeybees (Connor, 1970; Crane, 1985; Bradbear, 2009) play a critical role in pollination to enhance food security. Recent studies in Kenya, Greenhouse and open field experiments, involving a variety of fruits and vegetables: Strawberry, Tomato, Bean, Sunflower, Cucumber, Sweet pepper and other quick flowering plants, which are bee-pollinated, showed significant positive increases in fruit and produce, as a result of pollination, hence supporting the Greenhouse technology and pollination intervention, for sustainable production. It was recommended that stingless and solidary bees be integrated in farming systems, complemented by honeybees, for pollination of strawberries, tomato, beans and other fruits and vegetables, to boost food and nutrition security, for improved incomes to enhance rural livelihoods, hence quick economic growth to a middle income economy, as stipulated in: Livestock and Beekeeping Policies (2010), Agriculture and Apiculture Strategic Plans (2012-2017; 2017-2022), Agricultural Sector Development Strategy (ASDS, 2009-2020), Kenya Climate Smart Agriculture Strategy (2017-2026), Kenya's Vision 2030, Kenyan Government's Big 4 agenda (2018) and specific Sustainable Development Goals of the United Nations (2017).

[P.10.213] Characteristics of the tomato fruit (Lycospersicon esculentum Miller) using native Bumblebees as pollinators in greenhouse

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In Uruguay the production of tomato (Lycopersicum esculentum Miller) in greenhouse conditions presents pollination issues that limit its yield. The use of *Bumblebee*s (Bombus spp.) as pollinators can help overcome this problem as they perform, buzzing pollination, a behavior that makes them excellent pollinators of Solanaceae and particularly tomato. The aim of this study was to evaluate the effect of the native *Bumblebee*s *Bombus atratus* on the characteristics of the American tomato (ELPIDA variety). An experience was made in Canelones department where the fruits from flowers pollinated by *Bumblebee*s and flowers not visited by insects were compared and two experiences in Salto similar to that of Canelones but including flowers treated with hormones. In the three greenhouses analyzed the visit of the *Bumblebee*s to the flowers increased the proportion of fruit set in 13 – 47 % in relation to the result obtained in flowers not visited by the insects. Also, the pollinator action of *Bumblebee*s significantly improved the weight, size and number of seeds compared to fruits obtained from flowers without access to pollinators, although this improvement was only recorded in the experience in Canelones and in one of three greenhouses from Salto. In one of the experiences of Salto, flowers pollinated by *Bumblebee*s gave rise to tomatoes of greater weight, size and number of seeds than those from plants treated with hormones. This study is the first in Uruguay to show the benefits of using native *Bumblebee*s to improve tomato production in greenhouses.

[P.10.214] Botanical species visited by Bumblebees Bombus atratus used for the pollination of red clover (Trifolium pratense)

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Red clover (Trifolium pratense) is the most common forage legume in Uruguay. However, the country's demand does not have to be met. Among the main causes that impede improving yields is the deficit pollination. Previous studies showing the use of native *Bumblebee*s *Bombus atratus* as a pollinator can yield very good seed yields. The recent availability of colonies of B. atratus through artificial breeding translates throughout the year allows the use of this pollinator at the optimum time of cultivation (December-January), two months before the natural colonies, reach the peak of population (March). To determine the production of red clover seeds using colonies of B. atratus in December 2019, they installed 16 colonies of B. atratus (Brometan, Argentina) in a 6 ha nursery of red clover (variety La Estanzuela 116). During the period of flowering of the red clover 7 samples were taken from the contents in the pots of the nests and then their botanical origin was analyzed by palynological techniques. The presence of red clover pollen varied between 19.3% and 52.1%, coinciding with the maximum income with the peak of flowering of the crop. The most competitive plant species were: Eucalyptus spp. (6.2-47.2%), white clover (Trifolium repens) (2.9 to 19%), palm trees (Arecaceae spp.) (18.5-41.2%), lotus (Lotus corniculatus) (0.8-11.1%) and a native myrtaceae (2.2 -17.0%). The first two resources are present in the last three registers. Four colonies that did not have access to sugar syrup available in an internal dispenser collected more nectar from red clover (54 ± 24%) than the remaining 12 colonies that have availability of this food (27 ± 23%). These results have been predominant in red clover to obtain information (pollinating its flowers), although the competition of other species in the supply of this resource is important.

[P.10.215] The effects of inefficient bee pollination in primary industries can have devastating results on our ability to successfully produce florally-derived food sources - but through natural pheromone usage

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The purpose of this product development is to aid in the efficiency and effectiveness of bees to successfully pollinate pre-determined

floral areas.
Our environment is being devated by climatic changes, heavy mono-culture promotion, inappropriate pesticide usage and many other reasons, making it harder each year for bees to pollinate. Through the observation and the development of naturally-occurring pheromone based products, this trend can be reversed and bees can be directed to the required flora for pollination.
This dramatically increases fruit and vegetable yields. Pheromones from the lava, when sprayed onto blooms, attracts honey bees and other insects, to collect pollens where most pheromones are located. This will additionally attract bees from other areas. Pheromones can attract bees form other sources and direct them to areas requiring concentration for pollination, and also attract additional pollinators. Such naturally-occurring chemicals can also placate bees from exterior disruptors and prevent bees form pollinating areas not requiring pollination. Acting as an 'organic magnetic source' bees can now help the world to pollinate more effectively, with few bees initially. Thereby greatly increasing floral yields in the short-term, and greatly boosting pollination in the longer time frame.

[P.10.216] Observation and forecasting service of nectaring in Slovenia

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Slovenia with its 20,251 square kilometers of land is one of the smaller countries in the world. Regardless of its size, it has a very diverse climate and flora, which allows for the production of many varieties of honey, among which the highly regarded dark forest honey stands out with its premium quality. In Slovenia, around 440,000 ha or 58.2% of the total area of the country is covered with forests. Different conifers, such as spruce, fir and pine, can be found in most of them.
As early as 1901, the beekeepers recognized these natural riches and they laid the foundations of the Observation and forecasting service (ONS), which still operates today. By observing the development of plants and nectaring agents, the ONS predicts pastures all across Slovenia. An analysis of the condition of the nectaring agents on tree species takes place throughout the year with different assessment methods. In the autumn, it monitors the number of eggs or larvae, which are prepared for the winter hibernation and in the spring time, it analyses their wintering success. At a later stage, when the agents are already active, it again monitors their number and the intensity of nectar secretion as well as determines the degree of the nectar's ripeness.
Measuring stations are very helpful for nectar monitoring, as they help us oversee daily returns in a particular area. At the beginning of the Service's activity, the data was sent using special report cards every three days. Today, with the development of telecommunications, this is done much easier and faster.
The measuring station is a little house in which a hive is installed and underneath it an automatic electronic scale is placed. Every evening, this scale provides us with information regarding the honey yield, that is accurate to the decagram. In addition, we also record the air temperature and humidity of the surrounding area. In the evening, all of the measuring data is published online and on an app. This way, beekeepers are informed in a timely manner about positive and negative yields in individual areas.

POSTER SESSION 20 08:30-18:00
POLLINATION AND BEE FLORA III POSTER AREA

[P.20.517] Introduction of Medicinal Plants used by honeybee in Iran

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Medicinal plants contribute greatly to increase the therapeutic properties of bee products such as honey, pollen, royal jelly, and propolis. Also medicinal plants contain significant quantities of antimicrobial substances. Then the knowledge of medicinal plants enables beekeepers to utilize them at the maximum level, so that they can harvest a good yield and quality of honey with medicinal importance and other bee products in addition to effective pollination, which enhances crop yields. In this study, Pollen and nectar plants used by honeybees were gathered and identified from different regions of Markazi province through direct observation. Observations were made during spring, summer, autumn seasons. These were based on nectar and pollen source as well as activities performed by honey bees on different flowers. Simultaneously, to supplement the data, the pollens of these plants were gathered and examined by Erdtman acetolysis. In this study, we analysed 56 species of medicinal plants were found to be used by honeybees. Some of the common and important medicinal plants are the species of Composite, Rosaceae, Labiatae and etc. We can expect that this will result is useful for bee keepers to identify the medicinal plants and is helpful to improve quality of honey and sustaining medicinal biodiversity.

[P.20.518] On the Floral Origins of Mexican Honeys

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Bio Mieles del SUreste SA de CV. Quality Department, Mérida, MEXICO

At present, the Honey International Markets, requires the guarantee of the botanical origins of honeys, particularly for labelling purposes. Sensorial work is useful for the recognition of different honeys origins, but melissopalinology is the recognized method to determine it. This knowledge is useful to explore the possibilities to find new niche markets through the characterization at a Batch level.
In this work Honey Batches of finished product (n=20) from 3 important beekeeping regions in Mexico (Southeast, Central Highlands and Pacific Coast) has been examined for pollen. The aim was to determine the floral origins. Every batch is a blending of honeys from the same regions, localities or groups of producers. Honey drums were chosen according to the production time, production area and sensory features keeping the characteristics per blooming.
The honeys from the Yucatan Peninsula accomplish with the typical pollen spectrum recognized as Yucatán Honey, represented for the following Plant families: Burseraceae, Anacardiaceae, Sapindaceae, Fabaceae, Asteraceae, Polygonaceae, Sapotaceae, Euphorbiacea, Myrtaceae, Ulmaceae and Malvaceae.
Two Honey types, Campanilla and Multiflora, from the West Pacific Region, have, Fabaceae, Asteraceae, Tiliaceae, Lamiaceae, Convolvulaceae as its major components.
From Central Highlands of Mexico, Acahual Honey has pollen mainly from the Asteraceae family, Multiflora honey have Burseraceae, Fabaceae, Brassicaceae Sapindaceae, and Myrtaceae as the most representative families for pollen.
For the Yucatan Península, Bursera simaruba, and Viguiera dentata may result in unifloral Batches (pollen > 45% represented). From West Pacific, the Campanilla Type Honey has sub-represented the pollen grains of Convolvulaceae in the batches of this origin. Acahual type honey from Central Mexican highlands has usually unifloral origins (Asteraceae: Biden's type honey).

[P.20.519] Identifying, sourcing, propagating & nurturing of native & non-native floral sources

K. Banerd
Garden By Design, Dundas, CANADA

As a beekeeper or a person with an interest in bees, you know what bees need, but do you know how to identify, source, and nurture those floral resources?
- What are some great sources (tree, shrub, perennial or annual) of pollen and nectar for bees?
- What floral sources are native to northeastern North America, when do they bloom, and how can you identify them?
- What blooms during the common summer dearth period (late July and August)?
- How can you increase floral sources to extend the nectar/pollen gathering season for honeybees?
- What floral sources can you propagate by seed (perennial and annual)?
- Where can you source and purchase native perennial plants or seeds.
- What are desirable growing conditions for valuable floral resources (growth zone, sun exposure & water preferences, & soil type)?
- Bee turf and wildflower meadows versus monocultured, manicured lawns

•How to promote biological diversity in urban and rural environments.
•How to establish wildflower meadows.

[P.20.520] From Bloom to Boom: An investigation of Monarda fistulosa var. menthifolia for potential bee and human health

M. Kirby [1], R. Heyduck [2], T. Bates [3], J. Evans [4], D. Hyder [5]
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Our goal as a team of professional farmers and researchers is to examine and promote Monarda spp. as a new crop or accessory planting to affect bee health in situ and also produce a hive product and field crop that can be processed in a number of ways either as honey; a dried herb (flowers and leaves), an extracted product containing the volatile compounds, and as a value-added product to support farmer and beekeeper entrepreneurship.
Monarda fistulosa var. menthifolia, is a widespread North American native plant (also known as bee-balm, bergamot, or oregano de la sierra) that possesses a similar chemical profile to oregano including carvacrol, thymol, alpha-pinene, beta-pinene, sabinene hydrate, alpha-terpinene, citronellyl acetate, and beta-caryophyllene (Zamureenko, et al., 1989). Specific to bee health, thymol has been used to successfully control Varroa mites and prevent fermentation and the growth of mold in bee colonies (Calderone, 1999), and thymol based formulations are already commercially available (Floris, 2004). In addition, essential oils of oregano have been tested as a supplement to realize the same effects (Sammataro, et al., 2009).
To analyze oregano de la sierra and its effects on bee health and potential human health benefits through value-added products, we collaborated with several additional institutions and laboratories both in state and through USDA-ARS. Our research evaluated Monarda as a habitat enhancing plant by assessing the presence and relative concentration of thymol and carvacrol in nectar, honey, and hive architecture while Monarda is flowering; and afterward, to determine the persistence of the chemical constituents and evaluate its effects on bee health. Objectives include analysis of Monarda nectar, honey, and pollen using gas chromatography to determine volatile compounds content and to determine Nosema and Varroa mite counts in hives with access to different diets.

[P.20.521] Honeybees in the Garden: A Dance with Evolution

G.S.C. Rice
HoneybeeLives, New York, USA

During the course of evolution, pollinators and plants have been involved in a seductive relationship that has been instrumental in creating the fecund world we live in today.
Angiosperms developed flowers which exuded nectar purely to lure pollinators past the reproductive components of the stamen, holding pollen, and the stigma receiving the pollen into the female, fruit/seed producing ovule of the flower. Guide marks to the nectaries were deployed as advertising, with nectar runways often highlighted by ultra-violet colors and/or scent guides.
Bees evolved as the best pollinators, with the Eusocial honeybee (*Apis mellifera*) providing the most reliable pollination for many plants. A colony of honeybees sustains itself over multiple generations, which thrives by collecting pollen and nectar to feed young brood and producing honey for times of dearth. Many flowers have evolved to produce more pollen than the plants require for pollination as an adaptation. Different species of plants produce pollen and nectar at different times of day to limit competition for pollinator attention.
As single-source pollinators, Honeybees engage their communication and navigation skills to enlist their hive mates to bring home the goods. Von Frisch's waggle dance experiments laid the framework for understanding the complexity, and accuracy of this communication. Further study has revealed that bees use pheromones not only for in-hive communication, but also use "footprint pheromone" left at the foraging site to communicate enticement or depletion.
Communication signals between the flowers and the pollinators also exist. The electro-magnetic field of flowers can be sensed by bees, and it changes once a bee has visited, acting as a safe guard against false advertising to the bees. Electro-magnetic attraction plays a part in pollination, as the positive charge of a flying honeybee interacts with the negative charge of the rooted flowers, causing pollen to jump onto the hairs of the bee, aiding in the transfer of pollen to another flower.
Insight into this critical relationship between flowers and honeybees will augment the joy of watching bees forage, with awareness and knowledge, and potentially improve choice of forage plantings.

[P.20.522] The main Iranian Monofloral honeys

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Honey is a valuable food or nutritional supplements that is the result of the staunch symbiosis of honey bees with flowers and plants. The high diversity of nectar resource has led to the production of unparalleled range of honey of various herbal origin and with abundant medicinal properties in Iran. Due to the high diversity of beeplants, about 60 percent of honey production in Iran is polyfloral and about

40% of these honeys are known as monofloral honey due to the dominance of the nectar of a particular herbaceous species. According to the official statistics of agriculture Jihad in 2017, in Iran, 88,000 tons honey from 7.5 million honey bee colonies has been harvested, which are often located far from chemical inputs in mountainous ranges and steppe areas. The appropriate ecological conditions and the richness of plant species producing nectar and pollen have been caused the iranian plateau to be home to two Apis species honey bee. small honey bee, *Apis florea* that played a significant contribution in pollinating of field crops in southern regions of Iran, and its honey was harvested and sold as wild colonies and *Apis mellifera* meda, that is a native of the Alborz mountains and central regions of Iran, and has a major contributor to Iran's honey production. According to field studies conducted in Iran, more than 40 types of honey with high production levels are harvested, the most important of which Presian Sidr (Scientific name: Ziziphus spina-christi),Gavan gazangabin (Astragalus adscendens), Linden (Tilia begonifolia), Sumac (Rhus coriaria), globethistle (Echinops ritrodes), Fennel (Foeniculum vulgare), Coriander (Coriandrum sativum), lucerne (Medicago sativa), Citrus (Citrus spp),Kalpooreh (Teucrium polium),Thyme (Thymus daenensis), Sunflower (Helianthus annus), Eucalyptus (Eucalyptus camaldulensis), Acacia (Robinia pseudoacacia), Chaste tree (Vitex agnus – castus), Barberry (Berberis vulgaris), Russian Sage (Perovskia abrotanoides), camelthorn (Alhagi persarum), Dill (Anethum graveolens), Jujube (Ziziphus jujube), Chavil (Ferulago angulate),Kahoor (Prosopis cineraria), cotton (Gossypium herbaceum),yellow Milkvetch (Astragalus microcephalus), thistle (Cirsium bracteosum), Spurges (Euphorbia sp.), Pichak (Cynanchum acutum), Persian oak honeydew (Quercus Brantii),Taranjabin Honeydew (Alhagi maurorum).

[P.20.523] Automated honey pollen counting and identification

T. Braggins [1], R.M. Hodgson [2], J. MacDuff [2], P. Patel [1]
[1] *Analytica Laboratories, Hamilton, NEW ZEALAND,* [2] *Veritaxa Ltd, Palmerston North, NEW ZEALAND*

Melissopalynology is used to determine the geographic origin, floral source, and authenticity of honey. The technique involves separation of pollen from the honey and observation of the pollen under a light microscope. Pollen grains are quantified and classified according to their plant species origin by a highly trained expert. Although the preparation of the pollen is straight forward, the classification and counting of pollens is very time-consuming and reliant on specialist expertise gained after considerable training and experience.
To improve sample throughput, reduce the reliance on specialist expertise, improve precision, and reduce the tedium experienced by microscope operators, we have investigated the use of an automated purpose-build dark-field lighting digital duel microscope system that incorporates a neural net classifier and algorithms to count and classify high resolution, full depth of field pollen images.
This presentation will describe the operating principles and performance of the Classifynder™ in a commercial laboratory context.

[P.20.524] Using pollen DNA metabarcoding to investigate floral visitation by honeybees

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The National Botanic Garden of Wales uses pollen DNA metabarcoding to investigate which plants honeybees and wild pollinators visit throughout the year and the extent to which these can be provided within gardens. Here the Botanic Garden with adjacent Nature Reserve and organic farmland have been used to assess foraging by honeybees in order to build a temporal and spatial picture of plant use. Each month, from April to September, all plants in flower throughout the study site were recorded and honey was sampled from six hives. Three of the hives were set within an apiary within the Botanic Garden with close range access to horticultural habitat, while the other three hives were placed 1 km away within the Nature Reserve. We used DNA metabarcoding to survey which plants honeybees use by assessing the pollen biodiversity within honey, using the DNA barcode regions rbcL and ITS2. Initial results show that only a small proportion of available flowering plants are visited by honeybees within a diverse landscape. In total, 136 plant taxa were found in the honey from April to September with rbcL, but only 16 of these plants represented more than 10% of the DNA sequences returned for each month. The greatest proportion of DNA comes from native plants, including Rubus fruticosus, Trifolium repens, Hedera helix, and Taraxacum officinale, supplemented with lower levels of horticultural species. To complement this work, we have also analysed 475 honey samples provided by beekeepers to survey which plants honeybees use on a wider scale, throughout the UK. We are using our findings to develop evidence-based guidance on plants for pollinators and are working with specialist plant nurseries to pilot a 'Plants for Pollinators Assurance Scheme'.

[P.20.525] Pollenic and physicochemical characterization of honeys from Casamance Senegal

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To contribute to the knowledge of the nectariferous and polliniferous plants used by *Apis mellifera* in Casamance, two methods of approach (pollenic and physicochemical analyses) are used to apprehend this unexplored field of knowledge in Senegal. Our first sketch of honey typology was done on the 40 samples. 23 species have been identified in 23 genera and 11 families. The most represented families are Fabaceae (9 species), followed by Malvaceae and Lamiaceae with 3 species each, followed by Arecaceae and Meliaceae with 2 species

each, and finally Rubiaceae, Acanthaceae, Anacardiaceae, Asteraceae, Chrysobalanaceae and Rutaceae with 1 species each.
Of the 40 spectra, 26 have a dominant taxon (FR> 45%). The only pollen type present in all the samples is Elaeis guinensis (100%). Asteraceae pollen has the highest FR 97, 25%. Considering the total gross quantified spectrum, the nectariferous taxa dominate with 44.83% followed by nectariferous and pollinating taxa with 37.93% and finally polleniferous taxa with 17, 24%. The dominant nectariferous taxa are Ceiba pentandra with a FR max of 62, 45%; Avicennia germinans (47.33%), Daniellia oliveri (50.75%), Combretaceae (86.71%) and Myrtaceae (61.93%). The accompanying nectariferous taxa are Pterocarpus erinaceus and Parkia biglobosa with respective frequencies of 39.58% and 28.56%. Western Anacardium and Carapa procera are found only in non-centrifuged honeys with RFs not exceeding 6%. The quantitative analysis of the spectra shows that the class III (absolute density for 10g of honey between 100000 and 500000 grains) is the most important with 14 spectra is 35%. All drained honeys are in this class III, most of the centrifuged honeys are in Classes I and II. Pressed honeys are found in class IV and V.
37 (or 92.5%) samples have a water content of less than 20%. The average electrical conductivity of the samples is 8.5 10-4 Siemens cm-1. The polyphenol level is between 72.5 mg / kg and 403.9 mg / kg. The PCA shows the positive correlation of water content and electrical conductivity on the one hand and phenolic compounds on the other, with respective person ceffiscients of (r = 0.428) and (r = 0.433).

[P.20.526] Floral nectar characteristics on selected individual of Robinia pseudoacacia L. in South Korea

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The purpose of this study is to compare and analyze the differences in nectar characteristics, free sugars and free amino acid contents of the selected individuals of the Robinia pseudoacacia L. (false acacia), which have been proven to be valuable honey source among the individual trees. The volume of the secreted nectar per flower during one days was 1.7 ul averagely. Secreted nectar per flower of 'Pink false acacia (PA)' was 3.0±1.1 ul, which is highest value of secreted nectar volume. On the other hand, 'false acacia selected in Hungary (HA)' is the least secreted in nectar about 0.8±0.3 ul. Nectar sugar (brix%) by portable sugar meter was in the range of 40.1~51.4% and averagely shows 44.8%. According to HPLC analysis of collected nectar, the free sugar content in 'Wild type false acacia (WA)' individuals was 78.3±1.1 ug/ul, 'HA' was 38.8±5.8 ug/ul. Statistical analysis (Kruskal-Walis H test) revealed significant difference in free sugar content between selected individuals. Sucrose/Hexose ratio was 3.5±0.9 averagely, which includes 76.9% of sucrose, 4.5% of glucose and 18.6% of fructose, respectively. The amount of sugar per flower was calculated by multiplying nectar quantity and free sugar content (ug/ul). The results showed that the highest value was 155.6 ug in PA and the lowest value was 30.4 ug in HA individuals. Asparagine, alanine, glutamine, serine and glutamic acid were found in 79% of the total amino acids, and asparagine(59.8%) was the most abundant amino acid.

[P.20.527] Foraging activity of honey bees (*Apis mellifera carnica* Poll. 1879) on mint (Mentha spp.) nectar flow

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Biodiversity of nectariferous plants from different climatic and geographical regions of Croatia (Pannonian, Mountain and Adriatic) provides a great potential for the production of specific (rare) unifloral honey types. However, investigation of the honey bees' foraging activity on many less represented nectariferous plant species have not yet been carried out. Therefore, the aim of this study was to investigate the behavior of honey bees on mint nectar flow by monitoring their foraging activity. The study was conducted on five experimental colonies of Carniolan bee (*Apis mellifera* carnica Pollmann, 1879) placed in standard Langstroth-Root (LR) hives situated at selected locations in the mint field (Croatia, Pannonian region). Foraging activity of the honey bee colonies was conducted by counting the foragers on five separate 1 m^2 plots on mint field for six days during August. Counting of the honey bees was carried out for 1 min on each side of the plot three times a day (at 10:00, 13:00, and 18:00). Analysis of the honey sac content and pollen loads was carried out according to the method of Soehngen and Jay (1974). Statistically significantly higher (p=0,008) number of foragers with nectar and pollen load was determined at foragers collected at 14:00 (nectar: n=632; pollen load: n= 424) compared to those collected at 9:00 (nectar: n=582; pollen load: n=397), and 18:00 (nectar: n=560; pollen load: n=369). Statistically lower (p<0,0001) average weight of the honey sac content (9.02 mg) was determined in the samples of foragers collected at 9:00 compared to samples collected at 14:00 (12.04 mg) and 18:00 (11.06 mg). The results have also revealed statistically significantly lower (p<0,0156) average pollen load weight (7.73 mg) in the group of foragers collected at 9:00 in comparison to those collected at 14:00 (8.63 mg). No statistically significant difference of the pollen lead weight was determined between the groups of foragers collected at 9:00 and 18:00, and between 14:00 and 18:00.

[P.20.528] Sugar concentration in the nectar of the Dombeya wallichii flower and in the honey crop of the Africanized worker honeybee

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This research was carried out to evaluate sugar concentration in nectar throughout the day, in Dombeya wallichii flower and in honey crop of Africanized worker honeybee. The sugar concentration of nectar in the flower and in honey crop were measured throughout the anthesis period by collecting 10 flowers and 10 worker honeybees per hour (8 am to 5 pm) for three days. The nectar was collected with a capillary (0.001mL) and the contents placed in refractometer (Atago Refractometer). The sugar concentration in ºBrix degrees differed significantly by the Tukey's test at p <5%, in which the content of the honey crop presented mean of 14.20%, higher than the 12.33% of sugars found in the nectar of the flower. However, there was no significant difference in sugar concentration in nectar of the flower as well as in the honey crop content between the evaluation schedules. The sugar concentration ranged from 9.70 to 15.70% in nectar of the flower and from 12.20 to 15.60% in the contents of the honey crop. The sugar concentration in flower nectar remained constant throughout the day, thus D. wallichii ensures a higher frequency of floral visitors, ensuring their reproductive effectiveness. In the same way in the honey crop of *A. mellifera*, in which the worker honeybee carries a vital resource throughout the day in periods that the natural resources are restricted. There was a significative difference between the sugar concentration in nectar collected from the honey crop in relation to nectar directly collected in the flower, probably due to the digestive process that suffers inside the honeybee digestive system reducing the amount of water and, therefore, increasing the sugar concentration of the nectar allowing the transportation of a larger volume of nectar at a time.

[P.20.529] Plant sources of propolis in Iran

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Due to the rich flora of plant species and various climates, Iran has a high diversity in gum and resin sources. According to the latest botanical reports, more than 8,000 native plant species and more than 3,000 non-native species are growing in Iran, of which more than 1000 species are very important in beekeeping in terms of sources of nectar, pollen, propolis and honeydew .On the Iranian plateau, based on climatic conditions, there are five vegetation regions of Turanian, Hyrcanian, Zagros, Omani Persian Gulf and Arasbaran, which are important in each of these vegetative regions of Indicator plant species in beekeeping and especially in the production of propolis. The propolis, or the bee glue, is mainly a viscous resin material that is collected by *Apis mellifera* from buds, leaves, epiderm, secretion material from various trees and plants, and then inside the beehive, it is processed by adding some wax and pollen. Propolis produced in Iran has different colors of yellow, green, dark brown and generally olive green. Among the various factors affecting the production of propolis, including geographical and climatic conditions, race and population of colony bees, and the amount of Food storage of beehive; Resin and gum plants in the region are the most important in the quantity and quality of Propolis produced by bees. Since the physical and chemical properties of propolis such as color, flavor and odor, effective ingredients and its medicinal properties are influenced by gum, resin and plant origin, accurate knowledge of Plant sources of gum and resin in each region is necessary.Among the most important species of gum and resin, that are effective in the production of Iranian Propolis, can be referred to Cupressaceae family such as Juniperus excelsa, Salicaceae plants especially Populus nigra, Betulaceae family such as Betula pendula and Alnus glutinosa and Apiaceae species like Ferula gumossa, Ferulago sp. And Dorema sp. The necessity of more field and laboratory investigations to identify Plant sources of propolis in Iran is necessary.

[P.20.530] Palinoflora with apicultural potential used by *Apis mellifera L.* (Apidae) in Ripária Woods under the influence of Caatinga, Piauí, Brazil

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The bee farm in the state of Piauí (Brazil) is located in caatinga vegetation, with the reproductive period of the plants and, consequently, the honey production in the rainy season. Thus, most swarms are exposed to long periods of off-season (6 to 8 months) due to drought. It is during these periods that the swarms decrease rapidly due to the decrease of the food supply in the field. In this sense, during the dry season in caatinga areas, some plant species flourish, presenting as an alternative source of bee pollen. The importance of these interactions is essential to support programs for biodiversity conservation. With the objective of analyzing the apiary palinoflora, an apiary was installed in a transitional vegetation area, with monthly collections of reproductive branches, which were carried out in the dry and rainy seasons for analysis of the pollen grain and exsicata production. The research was carried out in a semi - arid region, close to riparian vegetation, with a predominance of buriti (Mauritia flexuosa L.), occurring transition to Caatinga, in the county of Santa Luz - PI, in the Ema locality. The area is located on the shores of the Corrégo das Emas, a tributary of the Gurguéia River. According to Köeppen's classification, the local climate is classified as Aw (tropical weather with dry season) with annual rainfall of 1010 mm and average minimum and maximum temperatures of 22 °C and 36 °C, respectively, with relative humidity of 62,3%. After being collected, the samples were placed in a greenhouse (60 ºC), inside wood presses for dehydration and incorporation into the Herbarium (TEPB) of the Federal University of Piauí. In addition, for the study of the palinoflora, the dehydrated pollen grains were acetolized, measured and described morphologically. A total of 52 species, 42 genera and 25 botanical families were found as probable sources of food resources, making three replication laminas of each of the 20 species studied so far. With this we have that the palinoflora is quite diversified, which implies in a high production of honey and derivatives, through favorable and drought periods, faced by A. melifera.

[P.20.531] Free sugar composition and amino acid content in floral nectar of prunus yedoensis Matsum and P. sargentii rehder

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The purpose of this study is to quantitatively confirm the value of honey source by analyzing the volume of secreted nectar, free sugar composition and amino acid content in floral nectar of Prunus yedoensis Matsum (Korean flowering cherry, KFC) and P. sargentii Rehder (Sargent's cherry, SC). While the flowering period of KFC was from April 6 to 15, that of SC was from April 10 to April 19. As a result of measuring volume of the secreted nectar per flower during two days by using 3 ul micro-capillary tube, the volume of nectar in KFC and SC was 7.6±2.8 ul and 8.45±3.3 ul, respectively. Because it was contained moisture in nectar, we conducted to dry the nectar at 80°C for 24 hours. The volume of dried nectar of KFC was 0.9±0.3 ul and the average moisture content ratio (v/v%) was 81.0%. The volume of dried nectar of SC was 0.95±0.37 ul and the average moisture content ratio was 88.8%. According to HPLC analysis of collected nectar (KFC: April 11 to April 13, SC: April 11 to Arpil 13), the free sugar content of nectar in KFC and SC show 47.9±13.0 and 25.5±101.7 ug/ul, respectively. Considering the amount of nectar per flower, the amount of sugar calculated from a flower of KFC was 362.7 ul/ug and a flower of SC was 215.5 ul/ug. Therefore, the value of producing honey of KFC is more productive than that of SC. The free amino acid content of KFC was 55.2 ug/ul and Glutamine, Proline, Asparagine, Glutamic acid and Arginine were the major components. In the case of SC, the free amino acid content was 25.1 ug/ul on average. Proline, Asparagine, Glutamine, Arginine and Glutamic acid were the major components in order. The above five amino acids accounted for more than 80.0% of the total amino acids. In this study, the nectar characteristics of KFC and SC were confirmed, and considering that KFC has more number of flower than SC per unit space, KFC tree is judged to have more honey-productive value.

[P.20.532] Antioxidant Capacity of Honey in different regions and seasons in Uruguay

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Honey contains important antioxidant compound content that protects cellular components from the harmful action of free radicals. Their components vary depending on the geographic location and vegetation. The aim of this work was to characterize the honeys of Uruguay from different regions by composition of trace elements, and antioxidant capacity, total polyphenols, and palynological profile. Samples of honey were obtained in two seasons, spring-summer (pv) and summer-autumn (vo), from three regions of protected areas, Quebrada de los Cuervos (Q), Valle del Lunarejo (VL) and Esteros de Farrapos (EF) respectively, and a fourth region Los Cerrillos region (C), which is associated with greater human activity. The trace elements were measured by FAAS (ug/g). The results obtained for Cu show significantly higher values in the C regions (1.785) and EF (1.706), respect to VL (0.850) and Q (0.849) respectively. The values obtained for Mn in region C (4,668) were significantly higher than Q (2,703), VL (1,931) and EF (1,628). For Mo, the Q region presented 1.060 being significantly different from C (0.090), VL (0.088) and EF (0.062). The pv season showed higher levels of Cu (1,258) and Mo (0,749) compared to season v (1,064) and (0,073) respectively. The content of total polyphenols (TPC) determined by Folin-Ciocalteu, of the C region gave higher content of TPC, 66,442 mg GAE / 100g being significantly different from Q (50,417 mg GAE / 100 g), EF (36,876 mg GAE / 100 g) and VL (35,526 mg GAE / 100 g). It was not found a significant difference per season. The antioxidant capacity determined by DPPH (µmoles TE / 100g honey), did not present any differences between regions and seasons. The palynological analysis showed that the Q, VL and EF regions were characterized as multifloral honeys. The region and / or season of the year, presents differences in the content of trace elements and presence of compounds with antioxidant activity.

[P.20.533] Characterization of honeys from four phytogeographic landscapes in Uruguay in different harvest seasons

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Apiculture in Uruguay is an activity which is present in the whole country. The annual production of honey is 12,000 tons and 95% is exported in bulk and without differentiation. In spite of being a small country, Uruguay has different landscape units, generated by both native and introduced vegetation, that grows in different soils, with different degrees of human intervention and climate conditions. Honey with different properties could be produced, and should be characterized according to the market demands. The aims of this research were: to characterize the honeys produced in four regions of the country through physiochemical and nutritional variables, to identify their botanical origin by a palynological analysis, and to monitor the presence of contaminants by analyzing the presence of glyphosate. The following were determined: protein (Kjendalh), humidity (%), pH and conductivity (20% w/v), colour (CieL*A*B*), macrominerals (FAAS) and sugar profile (HPLC). Three of the areas that were analyzed are protected areas: Quebrada de los Cuervos (east), Valle del Lunarejo (north), and Esteros de Farrapo (west). The fourth area, Cerrillos (south), is an area associated to fruit and vegetable crops and agriculture, and it is close to populated areas. Altogether 66 honeys were found throughout the year. Some differences in the variables colour, protein, conductivity and mineral content were found, depending on the region and the season. Monofloral honeys for European species as well as for native species, have been found. Some floral species present conductivity levels which exceed the limit established by the European

norm; the content of minerals by the fertility in some of the areas, could explain this fact. The possibility of generating our own local limit so as to differentiate between honeys and honeydews, is proposed. The detection of exotic botanical species within protected areas and minor traces of glyphosate, confirms the capacity of environmental sentinel of *Apis mellifera*, and could instruct government agencies with reference to the geometry and size these areas should have.

[P.20.534] Investigating in the bee shelters through bees and associated insects diversity and melliferous plants potential at a lime quarry in the Republic of Benin

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The bees play one of the most important ecological functions on earth and improve livelihoods. Unfortunately they are threatened worldwide and some of them are more confined to restricted areas such as quarries as their last shelter. In order to evaluate the contribution of the lime quarry of Fongba to biodiversity conservation and pollination service, the established bee populations at the quarry and their potential food sources were studied from February to August 2018. The bee diversity was studied by combining transect lines, hand-nest cashing and trapping methods while the melliferous plants were accessed through vegetation. A total of 44 plant species of 15 Families and 41 genera were recorded and most were concentrated at the extraction area. Seven of the 8 surveyed trees were melliferous while 30 of the 36 herbaceous species were melliferous. The melliferous potential of the quarry is essentially made up of herbaceous (74%) dominated by Tridax procumbens (14%). This particular area in the global floristically degraded district of Lokossa is host to 8 bee species, 6 pollinator fly species, 4 pollinator wasps and 1 predator wasp species. Dealing with the bee, 2 social bee species (*Apis mellifera adansonii*, and Hypo*Trigona* ruspolii) and 6 solitary bees (Seladonia jucunda, Pachynomia amoenula Chalicodoma cincta Eutricharaea sp., Xylocopa luteola and Xylocopa nigrita) were found. Seladonia jucunda was the most frequent (25%) in the site followed by Hypo*Trigona* ruspolii (20%) and *Apis mellifera adansonii* (20%). 56% of the insect density was concentrated at the extraction area and 81.1% of them were bees. As far as the social bees were concerned in particular, 5 *Apis mellifera adansonii* and 6 Hypo*Trigona* ruspolii colonies were found at the extraction area where they were established in human facilities and create many discomfort to the staff. A bee bank was created at the quarry to host the social bee swarms and the solitary bees. It proved to be a sustainable solution to the accidents that emerges from the social bee invasion at industrial quarries and constitute potential additional incomes to the community for poverty alleviation and biodiversity conservation.

[P.20.535] Soil microorganisms improve pollination landscapes

V. Strogolov
Strong Microbials, Inc, Milwaukee, USA

Pesticide and herbicide use can disturb microbial ecology of the soil, affecting mycorrhizal symbionts of plants. Successful pollination landscapes depend on strong plant mycorrhizal networks. Restoration of soil can take advantage of microbial remediation, which can facilitate decontamination and have added benefits of adding to the microbial and mycorrhizal community of the soil biome. Efficiency and density of mycorrhizal associations with plant roots mediated by soil, seed, and root inoculation is evaluated from the perspective of bee habitat quality. A case study comparing the effects of soil, seed, and root mycorrhizal inoculation on germination, growth, and productivity of flowering plants is addressed in this work.

[P.20.536] Conservation of Forage and Habitat for Bees in Uganda

J. Ssettaba
Native products ltd, Kampala, UGANDA

Bees collect pollen from flowering plants as a source of proteins, fats and other micro-nutrients. Despite the critical role pollination plays in Agriculture, Uganda is witnessing a substantial decline in the abundance and diversity of domestic and wild pollinators, including bee foraging species in Uganda, due to weak apiculture pollination policies.
The method of providing pollination services to bees (indigenous plants) has not been put into practice, beekeepers still depend on wild plants, forests as the major source of forage resources for their bees.
Bee pollination provide critical ecosystem system services vital to the production of numerous crops in the Ugandan agricultural sector ,An estimated 60 to 90 % of wild plants and 35 % of global crop production depends on animal pollination according to the information collected by patterns of pollinator biodiversity and economic values of pollination services in Uganda. Most bee colonies in Uganda are affected by habitat losses due to increased activities carried out like bush burning ,deforestation ,charcoal burning ,spraying using other chemicals which bees and wild crops .
Uganda grows more than 80% crops that require pollination to their fruits and seed. With increasing agriculture intensification, there is a high risk of pollinator depletion which might lead to failure of crops.
The abundance and presence of biodiversity on agricultural land like bees and other organs are important for maintain the healthy and productivity of farm lands

[P.20.537] Pesticides and Pollinators: Policy, Practice, Activism

P. Algara
BASE Landscape Architecture, San Francisco, USA, With Honey In The Heart, San Francisco, USA

The decline towards extinction of countless insect species is on the rise due to habitat loss, pesticide use, the proliferation of parasites and climate change. Rural lands dominated by monocultures are food deserts for pollinators, making it impossible for bees to survive throughout the seasons.
We believe the green areas in the urban environment represent a unique form of green infrastructure. Neglected sites along transit routes and waterways can be converted to trails of constant blooms that provide for diverse migratory and native pollinators.
We are currently working with the San Francisco Department of the Environment and the SF Department of public works on a demonstration site (a median on a busy intersection) to quantify the increase in bee presence and diversity as well as mapping the city's pollinator gardens to create networks of connections. The garden uses only non treated plants as well drought tolerant and our planting palette provides year round source of nectar and pollen. We did a comparative bee count and found 11 species on the planted median versus one on the non-planted medians. If replicated in other places, migrating pollinators could replenish themselves on a city-to-city nectar circuit and native species could have a healthy habitat to thrive.
Increased awareness about the importance of designed pollinator habitat has unfortunately not been matched by frank discussions about the risks of systemic pesticides in nursery-grown plants that are used in the design of landscapes. In this session we will present topics around pesticides & pollinators and learn how growers, policy-makers, and urban planers and landscape designers can work together to create change at multiple levels in the urban environment.

[P.20.538] Borders in crops, designed to support pollinators

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There is a growing loss and modification of insect populations in the world, associated with the modification of environments by the cultivation of soils. We also have an increasing demand for food, and many of them depend heavily on insects pollinators. It's therefore important to conserve spaces where pollinators can obtain food, build nests, obtain shelter and have safe mating sites. In this way it contributes to the conservation of insects pollinators.
Two commercial farms were selected in the Department of Soriano-Uruguay, with soil management for traditional crops (Prairie, Sorghum and Soy), with the aim of evaluating some edges of crops, as a multifunctional landscape of interest for pollinators. In each farm an area contiguous to the crop was conserved, of 10mt of length x 2mt of width that remained without tillage. Periodic survey of plant species and insects pollinators, present in these areas, was carried out. The insects were recorded by direct observation on the flowers and with a water trap (yellow) with soapy liquid.
As main results we obtained that: 1 - In the excluded areas, plant species of food interest arose for pollinators (isolates): Trifolium repens, Cichorium intybus, Taraxacum officinale, Echium plantagineum and Eryngium horridum among others. 2- The presence of grasses in these reserved areas depletes the outcrop of the dicotyledons 3- The richness of pollinating species is greater in the months of December / January. The greatest diversity of species was registered in the group of bees, with 14 species of native bees and *Apis mellifera*, present in these selected areas (captured in water traps). 4 - The importance of Eryngium horridum as support for the mating of a species of native bee of the Halictidae family. 5-These reserve areas contribute to the conservation of pollinators, especially bees. A control of monocotyledons (Poaceae) is suggested before reserving the exclusion areas to allow the upwelling of the dicotyledons that are preferred by insects pollinators and to add structures that serve for the installation of their nests.

BEEKEEPING TECHNOLOGY AND QUALITY

10 SEPTEMBER 2019

POSTER SESSION 11 08:30-18:00

BEEKEEPING TECHNOLOGY AND QUALITY I POSTER AREA

[P.11.217] Real time brood thermoregulation detection

E. Henry, X. de Briey, M.-A. Roberge
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Honeybee larvae and pupae depend on accurate regulation of brood nest temperature between 32–36°C for proper development. Worker bees regulate brood nest temperature through metabolic heat. Interruptions in thermo-regulation can be caused by important events throughout the beekeeping season, like entering winterization, or breaks in the egg laying cycle.
Temperature and humidity Nectar sensors were installed in the brood super of 96 beehives in the summer of 2017 in 5 locations in the greater Montreal area. Data was collected every 30 minutes until November 2017, and each hive sensor sent data to the cloud over 3G.
To detect thermo-regulation states is in real-time, a temperature derived variable was used to create classes that represented the thermo-regulation and thermo-regulation-less periods. A 24-hr moving average of the standard deviation of the temperature was computed for each beehive, then grouped into thermo-regulation periods if less than 4 deg, and thermo-regulation-less if greater than 4 deg (the difference between 36 and 32 deg C) With the data labeled by class, the data was split into 80% 20% testing and training data, and k-group fold cross validation was used preserve the ratio of the classes in the training and testing data
The resulting algorithm provided a mean accuracy score of 96.8%, a precision of .991, a recall of .971, Specificity .957, and F1 Score .981. In five instances during the summer of 2017, Nectar alerted beekeepers when beehives stopped thermo-regulating, and 100% of the time the queen was absent during the thermo-regulation-less periods.
Real-time detection of thermo-regulation of brood can help beekeepers take better management decisions regarding queen replacement during the season and winterization treatments in the fall. Future data collection will contribute to improving the detection time as well and understanding dynamics of thermo-regulation in general. Future research on understanding the relationship between thermo-regulation states and queen health will provide more direct information on queen health.

[P.11.218] Data Mining to Understand the Benefits of Good Beekeeping Practices

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All around the world there are many standard practices for beekeeping. Some have developed from the use of natural materials and ancestral traditions that are typical of the different geographical areas, while others depend upon scientific research. As pollinators, honey bees alongside with wild pollinators support biodiversity of wild plants and contribute to increase yields of important highly valued agricultural crops. Modern beekeeping is facing numerous challenges due to a variety of factors, mainly related to globalization, agrochemical pollution and environmental changes.
A list of Good Beekeeping Practices (GBPs) was developed by the Istituto Zooprofilattico Sperimentale del Lazio e della Toscana, along with other leading scientists and beekeepers, in the context of a specific EU project (http://www.izslt.it/bpractices/) on the basis of creating a list of sustainable and resilient beekeeping methods. Daily implementation of the GBPs in apiary management would result in beneficial impact on several aspects: economical (cost reduction, larger production per unit, higher income for beekeepers), on beekeepers safety (safer handling of veterinary medicines), on consumers health (better quality of honeybee products preferring treatment with low environmental impact products, lower residual levels in honeybee products due to a safer use of medicines), honeybee health (better preventative measures, higher application and efficacy of beekeeping techniques and control methods, higher performances) and environment protection (proper management of smoker, use of organic treatments, biodiversity preservation).
A proper honeybee colonies management should consider a wider vision, in a “one health” approach in order to protect honeybees, humans and the environment. In order to achieve those objectives, we attempt to validate key GBPs via data mining techniques and statistical practices. This study intends to justify GBPs, through the process of definition, validation and classification done in the creation of the list; and through data mining identifying GBPs critical benchmarks using software records and evaluation through data analysis methods. We will use apiary management software data to verify the use of record-keeping, disease inspecting, providing treatments, monitoring queens, and other GBPs in the promotion of a more wide approach to the health and well-being of the hives.

[P.11.219] Automated Varroa counting system on sticky boards based on image recognition

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Varroa causes numerous damages to bee colonies: deficit of production, virus development and winter losses. Treatments are essential and recommendations in northern hemisphere are : before winter, remove 80% of Varroa mites from colonies to limit mortality; in winter, reduce the number of Varroa mites below 50 individuals. Hence there is a need to monitor the population dynamics of Varroa.
Currently no measurement system enables to know exactly the population of mites in a colony, but some methods can statistically approximate the number of phoretic mites. The natural fall method offers the advantage of being non-destructive for bees and non-disturbing (no need to open the hive). It consists of placing a sticky board (usually coated with oil, wax or glue) at the bottom of the hive beneath a grid to trap the Varroa mites which fall naturally. By manually counting the number of mites collected on the entire board, their number reveals the infestation rate of the colony.
However, the manual comprehensive counting method is tedious, boring and causes unpredictable random errors. The automatic Varroa counting method presented here is an innovative tool based on deep learning and image recognition: algorithms are supervised to recognize mites on a board cluttered with other residues. The learning set is provided by a large database of images where Varroa mites have been manually tagged. The resulting set of manual labels serves as a reference for evaluating the accuracy of the automatic counting method (a linear regression compares the results of manual counting and automatic counting). The error estimate is given by false negatives (Varroa mites missed) and false positives (Varroa mites when it should not be). The precision of the results varies according to certain variables: density of residues on board, total number of Varroa and resolution of the images. This method offers solutions to the current need to standardize Varroa counting in order to control the effectiveness of treatments and the Varroa population. The whole method and the first experimental results are presented.

[P.11.220] Communicating with your hive using the Internet of Things. Connecting hivescales via LoRaWAN and The Things Network

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LoRA (Long Range) wireless transmission offers significant advantages for Internet of Things (IOT) communication compared to WiFi, Bluetooth, cellular or satellite transmission, since it provides greater range whilst requiring less bandwidth and power. Electronic scales transmitting beehive weight are a typical application.
The Things Network (TTN) is an open source project that uses LoRaWAN (wide area network) wireless transmission to connect inexpensive low power devices with network gateways. These gateways transfer the data into the 'Internet cloud'. The many existing TTN gateways are available for any user to utilize. Alternatively, a low cost gateway can be purchased and connected to the Internet. Each gateway can service hundreds of nodes within its range. The notional range for the link between node and gateway is up to 7 km depending on aerial configuration and line of site.
The hive scale described here consists of 4 x 50Kg load cells monitored using a Sodaq Explorer (Arduino) microcomputer furnished with a HX711 amplifier. The board features an integral RN2903 LoRa radio module. The board is put into a low current consumption 'sleep' mode between measurements and the LiPo battery charged by a small solar panel allows it to run indefinitely.
The module connects to a TTN gateway with 'over the air activation' and data is forwarded to ThingSpeak for storage and display. The device was configured to wake and take a reading every hour. Over the course of 5 weeks there were approximately 700 readings, recording an increase in total hive weight of 8.8 kg.
LoRaWAN communication is reliable and affordable. It has the potential to offer a cost-effective solution in remote areas where cellular networks are unavailable, as data from a large number of hives within a few km radius can be collected by a gateway connected to the Internet with a satellite modem.

[P.11.221] ApisProtect

F. Edwards Murphy
ApisProtect, Cork, IRELAND

Dr Fiona Edwards Murphy is the CEO and co- founder of ApisProtect Limited.
ApisProtect provides an in-hive sensor network with long-range, cellular and satellite-powered communication to proactively monitor honey bee colonies. Combining the sensor data on hive conditions, health and activity levels with its proprietary big data and machine learning techniques, ApisProtect gives beekeepers actionable insights and alerts to help prevent losses and increase colony productivity. Beekeepers no longer need to rely solely on periodic, manual hive checks that can allow disease, pests and other issues to deteriorate hive health beyond rescue.
Now monitoring the health of over ten million honey bees in hives across Europe and North America, ApisProtect brings the power of advanced sensors and machine learning technology into the hive to deliver a 24/7 early warning system so beekeepers can give at-risk

hives immediate attention and improve bee health.
Dr Fiona Edwards Murphy completed her PhD in October 2017 with the School of Engineering, and the School of Biological, Earth and Environmental Sciences at University College Cork in the area of Internet of Things applications for honey bee health.
Fiona is a 2013 graduate of the Bachelor of Electrical and Electronic Engineering (BE) Honours at UCC, a recipient of the Google Women Techmaker Scholarship 2016, and the Irish Research Council Government of Ireland Postgraduate Scholarship 2014. She was recently awarded the Women Mean Business Sodexo Newcomer of the Year Award 2018.
She placed in the top three of the 2016 "Ireland's Best Young Entrepreneur" competition in the "Best New Idea" category and was awarded "Young Entrepreneur of the year" at the U magazine 30 under 30 awards 2017.
She placed second in the Blackstone/Techstars global Student Entrepreneurs awards, and represented Ireland at the European Union 'Ideas from Europe" challenge, placing 2nd in The Hague in April 2018.
Dr. Edwards Murphy's work on the topic of hive monitoring has received many national and international awards from the Irish Research Council, The IEEE, IBM, The Irish Laboratory Awards, Google, and the Global Entrepreneurship Summit.

[P.11.222] sBee – Towards an Effective Beekeeping

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As happened with many other activities, the emphasis on ICT has proven to enable new paradigms and new approaches to overcome difficulties and to emerge innovative useful services.
Beekeeping is much more than a beekeeper/honey relation. The activity itself is ancestral with not so much new to learn, however, the way it is managed and maintained can surely be improved. The fact that it is an activity mostly supported by older and conservative people, make that traditional methods does not fit easily into the changing or integration of new tools or paradigms, such as information technologies.
Beekeeping is not only about managing hives for the production of honey, but all its adjacent and related activities: the efficiency on managing and planning interventions, the continuous beehives monitoring and sensing, the knowledge and application of rules and legislation, the agility to face to unexpected situations (diseases, fires, thefts, etc.), the efficiency on in-loco technical support, the traceability of the quality of the production, the research for health care and innovation with universities.
The sBee – Smart Beekeeping is an applied research project integrated on the Portuguese initiative to develop a set of new tools to improve the capacity and quality of the national beekeeping activity, based on: i) a distributed and integrated ICT platform for the national beekeepers ii) a federated social network to support the interoperability of all beekeeping stakeholders (beekeepers, associations, authorities, insurance companies, suppliers, clients, others); iii) electronic devices for remote monitoring and beehives ecosystems sensing and apiaries surround areas; iv) a set of innovative services to support decision-making, predicting bee behavior, anticipate corrective measures; facilitate the management of interventions; traceability of products.
The platform will be prepared for artificial intelligence services integration suitable to help improving effectiveness on beekeeping.

[P.11.223] Comparison between traditional and industrial production of caucasian honey bee queen

S. Rezaie Kangar Shahi
Pishroo Beekeeping Company, Tehran, Iran, kermanshah, IRAN

This study was conducted to compare the Caucasian honey bee queen production methods at a completely randomized design with 4 treatments and 8 replicates. Treatment were included of: 1) wet grafting method, 2) dry grafting method, 3) bow-like frame method and 4) traditional method as emergency queen production. Starter and finisher hives were fed syrup containing pollen. Queen cell height, queen weight at birth, duration between birth of queen to laying, laying queen weight and laying area during the first week were recorded as indices of queen quality and quantity. Based on the results, queen cell height, origin queen weight and laying queen weight were affected by the queen rearing methods ($P<0.05$), but duration between birth of queen to laying was not affected ($P>0.05$). The queen cell height in dry and wet grafting methods was higher than two other methods, but there was no difference between dry and wet grafting methods ($P>0.05$). The weight of origin and laying queen in dry and wet grafting methods was higher than traditional methods. There was no difference between both method and swarming method for origin queen weight ($P>0.05$), but the weight of laying queen in both method was higher than swarming method ($P<0.05$). There were no differences between dry, wet grafting method and both method for laying area, but difference exist with swarming method ($P<0.05$). Based upon these results, the application of dry and wet grafting methods could rear queens with the highest quality.

[P.11.224] Impact of Technology on Commercial Beekeeping

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Very few human beings on the planet are out of the reach of technology, and we all experience the relentless pace in which our personal

and business lives are being transformed by digital technology: introducing new ways of communicating, unprecedented data collection, and new software tools for increasing efficiency through data driven decision making. And although the beekeeping sector lags behind other agricultural areas in regards to the development, adoption, and integration of technology based tools, commercial beekeeping is beginning to feel the influence of this movement in Ag Technology.

We survey the current state of software and hardware solutions available to the commercial beekeeping industry based on ten years of experience with the Bee Informed Partnership in the United States and with Hive Tracks. Apiary management software, hive monitoring through a variety of sensors, hive and super identification, Varroa, nosema, and virus sampling data, and secondary digital data sources offer potential economic value to a commercial beekeeping operation, but factors such as cost, reliability, and ease of use influence the adoption of these technologies. Each of these factors will be examined to provide a practical framework for evaluating technology feasibility based on the specific circumstances and needs of a commercial beekeeping operation. Case study examples will be given of the experiences of current commercial beekeeping operations adopting these technology solutions and how they handle the challenges and obstacles of integrating technology into the fast paced and often chaotic commercial beekeeping workflow.

While digital technologies directly impact commercial beekeeping operations internally, a number of new economic opportunities are being created at the intersection of beekeeping operations and various stakeholders including honey buyers, pollinator dependent growers, government agencies, and survey takers. A beekeeping operation supported by data can leverage that information to command higher honey and pollination contract prices, meet regulatory and compliance requirements, and participate in ongoing research and development projects. In addition, reliable and trusted beekeeping data will foster the development of new insurance products and smart contracting options that foster commercial beekeeping business security and continuity.

[P.11.225] Potential of backside operating hives in future beekeeping

J. Bozic

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Beginning of twentieth century was time of spread of modern beekeeping technology with movable combs. In Europe were many different solutions and across the Ocean dominated Langstroth hive. Until nowadays, most of the European countries have established beekeeping that is based on principals of Langstroth hive with different frame size or hive sections arrangement. Only few countries have a long side also backside operating hives and only in Slovenia is dominated so called Alberti-Znidersic or AZ hive. At first view, it seems that traditional and cultural aspects of Slovenian beekeeping are the main reason. Many professional beekeepers are showing that, with some modification, such type of beekeeping could be profitable and at list competitive with use of Langstroth type of hives. Many operations inside of the hive are optimized and reduced with experienced beekeeper. Investment costs are reduced due to new way of hive construction. Construction of movable bee houses reduces and simplify costs of movable beekeeping and more efficient exploitation of honey sources, especially sorted honey. All existing modifications and solutions are compared and evaluated. Modern AZ type of beekeeping simplify pollination services, inclusion of electronics to monitor bee colonies and have potential for introduction of some automated processes in bee colonies management. It has been already shown to provide great solutions for apituristic services, especially special wellness programs in bee house. It also enables beekeeping to physically impaired people. With some future adaptations to the most spread size of the frame it could have wider potential around the word.

[P.11.226] The effect of traps' material and purification methods on the quality of propolis

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The effects of different purification methods on the qualitative characteristics of propolis extracts, and the effects of the construction material of commercial propolis traps (low-density polyethylene -LDPE, high-density polyethylene -HDPE or Polyvinyl chloride -PVC) on the quality of collected propolis were investigated.

Propolis collected from commercial traps was analysed with LC/MS for possible migration of trap construction material into the sample. No migration was detected with LDPE traps, but traces of PVC and HDPE were found in all samples from the PVC and HDPE traps.

For the purification of propolis, five different methods were deployed for ethanolic and methanolic extracts: a) centrifugation for 30 min at 370g; b) centrifugation at -5 C for 10 min at 1850g twice with a 15 min interval; c) centrifugation at 3340g for 2.5min twice with a 15 min interval; d) filtration through a 0.22μm polyethersulfone membrane; e) filtration through a 0.45um pore size nylon membrane filter using Buchner vacuum system.

The results using each method were evaluated with reference to the post-treatment antioxidant activity (AA), total phenolic content (TPC) and total flavonoid content (TFC) of each extract. Two methods of centrifugation were found to be the most effective and stable: centrifugation at 3340g for 2.5 min, and for 10 min at 1850g – both at -5C. Using the second method, there were increases of approximately 80% and 50% for the AA of ethanolic and methanolic extracts, respectively. Using the first method, there were increases of approximately 170% and 150% for the TPC of ethanolic and methanolic extract respectively, while the TFC increase was over 100% for both ethanolic and methanolic extracts.

[P.11.228] Beeswax foundation as a new method of transporting medical substances for bee disease control

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The control of known bee diseases – bacteria, bacilli, microbes, streptococci, fungi by way of existing methods is ineffective. We have offered a completely new method, not in use before, in treating and preventing bee diseases. It is based on restoring the protection against pathogenic microflora to the bees, which they would provide in nature for themselves. For the first time in the world practice of beekeeping we have proposed a mechanism of delivering/transporting the protection components to a bee family. It is worth noting that the therapeutic and preventive substance is introduced into the wax foundation to perform several important functions. For treating and preventing a number of diseases, of basic importance are:
- time of starting the treatment or its preventing
- time (period) of pathogenic microflora exposure to the drug
- a reliable and simple method of feeding the medical substance, ensuring a constant contact of a bee with the medical substance from an egg to adult.

[P.11.229] Development of an integrated system for processing of bee-pollen in high mountain areas from south american andes

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The area of the Cundiboyacense highland located in the central Andean region of Colombia, located at 2500 meters above sea level, is one of the zones with the highest production of bee-pollen in the world with yields close to 35 kg/hive per year. Bee pollen from this area has notable nutritional properties and compounds with functional properties such as phenols and carotenoids. These properties are given by the botanical sources of the zone rich in nutrients and secondary metabolites due to the geographical and climatic characteristics of the region. In contrast to these production advantages, the Cundiboyacense region does not have adequate processing systems for bee pollen, which leads to deficiencies in the quality and safety of the product. The improving of bee pollen processing facilities by technical modifications and the implementation of an integrated cleaning and drying system for the product in an apiary in the area was proposed. The beehives were modified through the installation of supports and alternative bee pollen traps to avoid sources of contamination. The processing facility was modified according to considerations of Good Manufacturing Practices with food processing technical considerations implementing the combined use of a cabin dryer with temperature control and forced air flow and a greenhouse-type solar drying system. Additionally, for the separation of impurities, a cyclone type system was implemented, complementary to screening equipment. With these modifications, a decrease in the content of impurities and the microbiological load of bee pollen was seen from the first stages, principally with a descend in molds and yeasts content and in the number of impurities (insects, stones and vegetal material). The use of the greenhouse solar dryer integrated to the cabin dryer allowed the processing of larger quantities of product with shorter waiting times in storage, reaching a moisture content and a water activity appropriate for the conservation of bee pollen. The contents of functional or nutritional compounds were not significantly altered, even observing an increase in phenols content and a not significant decrease in carotenoids content and antioxidant activity.

[P.11.230] Properties, production technology and efficient use of beeswax foundation with the addition of propolis (bee glue)

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Vital functions of honeybees depend on many internal and external factors, including the state of the brood nests. The quality of raw beeswax for producing foundation and the honeycombs rebuilt on it has a subsequent effect on the processes associated with the development, productivity and spreading of diseases in the brood nests. We have set the goal to ground theoretically and experimentally the efficiency of applying the additives of bee glue fractions in producing the wax foundations, as well as to improve the equipment and technology of their production and study the efficacy of their application.
Microbiological studies have shown that the wax foundation with additives of the bee glue fractions inhibits the growth of microorganisms during the first five days, including Ascosphera apis (by 85-95%) and Aspergillius niger (by 75-90%) and increases the survivability of bee-families in winter, when the reduction of bee wastes in winter due to a decrease of fecal load of colon and absence of excrement traces in the nest are observed.

[P.11.231] Apiculture and geobiology

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Humans have always had the tendency to orient things towards practicality in the most basic sense: the simplest physical routes, the easiest set-ups, thought-mitigating systems to create unthinking pathways. However, by focusing on this accessibility more than anything, we have filled our society with things which forget the flows and energies in nature, replacing them instead with material and processes which help us above all else. In apiculture this can be particularly destructive, as the sensitivities of the living hive are still a mystery to most people. It is therefore crucial to learn the intrinsic nuances of the spaces and material used for beekeeping, to better understand how to provide a full strength support for each colony. Catherine Ballot-Flurin and Olivier Raud will present their work on hive and wood polarity, energetic positioning and environmental flow, and how it relates to conscientious beekeeping. This will include the presentation of our polarized hive and the data have collected on the beneficial effects on the well-being of the colony.

[P.11.232] Inclusion of bee pollen oil extracts and Acacia mangium honey in oat cookies: carotenoid-enrichment and sensory acceptance

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High-Andean bee pollen has been found to be particularly rich in bioactive compounds, such as carotenoids, in comparison with pollens from other origins, whereas A. mangium honey has been proposed as a remarkable natural sweetener. There is little knowledge on their inclusion in processed foods. In this work, we explored the replacement of conventional fats (CF) by sunflower oil-extracts of bee pollen (BPE), and of sugar by A. mangium honey, in traditional oat cookies. First, formulations of oat cookies were prepared varying the ratio of CF:BPE (1.0:0.0; 0.5:0.5; 0.3:0.7; 0.0:1.0) at the same conditions. Addition of BEP produced cookies with a characteristic yellow hue and, as expected, an increased concentration of carotenoids (up to 33.8 µg eq beta-carotene/g). A consumer acceptance test, by 25 frequent cookie consumers, in a five-point hedonic scale, was performed to determine the acceptability of the product depending on the degree of CF replacement; the results showed that, despite the noticeable sensory differences regarding color and flavor, BPE inclusion did not affect overall acceptability. An analysis of convenience and utility considering carotenoid-enrichment, sensory acceptance and cost, allowed to select 70% substitution as the better cookie formulation. In parallel, two formulations of oat cookie were prepared using as sweetener: only refined sugar, and 50% sugar-50% honey, and subjected to a consumer test. Results showed that honey favorably increased the crust color and texture; although its inclusion significantly decreased the flavor acceptance. Finally, a formulation of oat cookie with 70% of substitution of CF by BPE, and 50% of sugar replacement, was prepared, subjected to 60-panellists acceptance test, and stored at room temperature in dark bags with low water permeability, during a month, to evaluate the stability of carotenoid content and color. Results confirmed that oat cookie with inclusion of bee pollen and honey from Acacia mangium, presented a high acceptance rate, with a stable yellow pigmentation and carotenoid content for at least 30 days. These results highlight the potential of using bee products as differentiated and functional ingredients of baked products, which ultimately add nutritional value and enable access to highly specialized markets.

[P.11.233] Impact of climate change on beekeeping practices

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CARI asbl, Louvain-la-Neuve, BELGIUM

The climate changes we are experiencing today around the world affect many aspects of our environment. The main focus is on the evolution of rising temperatures and extreme climatic events that have an impact on many elements including the loss of biodiversity, the evolution of flowering, changes, the biological cycle of bee colonies, the development of certain pathogens, the survival capacities of colonies, honey produced and its characteristics... The number of publications reporting its various points is constantly increasing. On the basis of a study of the main changes already recorded and the simulations available, various recommendations will be formulated in relation to good beekeeping practices in order to help beekeepers to better manage the maintenance of their livestock and production conditions. Concrete advice will be presented in relation to honey management, feeding, water requirements, and the management of certain pathogens. Solutions for settlement monitoring and training will also be formulated. The impact on the market of hive products will also be analysed.

[P.11.234] Multipurpose dispenser for bee hives (MDBH) - a new device for efficient and safe delivery of multiple formulations of acaricides/medicines to manage multiple pests in bee hives

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Commercial apiculture worldwide faces multiple biotic threats necessitating repeated application of pesticides that have wider bee, human health and trade implications. Organic management strategies use safer formulations through dispensers/devices that are pest specific,

costly, non reusable, adds to inventory problems impeding large scale adoption. To overcome such hurdles, a multipurpose dispenser for bee hives (MDBH) was designed using non corrosive, food grade molded plastic. MDBH consists of a solid base unit to hold chemical formulations. Top lid has a soft notch for delivery of chemicals and cross rows of perforations to allow its dissemination. It fits onto base unit. Base unit comes with a replaceable absorbent pad to hold chemical for sustained release. Sleek design allows multiple placement options at bottom board or on top bars, leaving sufficient space for free movement of worker bees inside hive. MDBH eliminates need of additional boxes and has universal application across bee hives including A. those of *A. cerana*. For liquid chemicals with single dose application, measured dose is poured directly onto absorbent pad through notch. After removing two brood frames form a colony, MDBH is placed on bottom board, aligning with rear wall of brood chamber. Colony is closed after replacing frames. For multiple dose formulations, initial loading operation is similar. Additionally, a straw is inserted into notch, its upper end popping up between two frames. Subsequent applications are made using a syringe through straw. Solid, powder, tablet or gel formulations are similarly placed in base unit after removing absorbent pad and refilling if needed is done. MDBH has been successfully used for multiple pests including Varroa with different formulations of formic acid, thymol (liquid, crystals, tablets) and many essential oils; endoparasitic mites with formic acid and methyl salycilate; wax moth with PDBC, aluminum phosphide, MB, etc. It has potential to be used against many other pests. Unique design ensures complete safety of worker bees avoiding direct contact with chemicals and quick delivery of chemicals. MDBH is cheap, reusable, easy to operate, clean and store. It not only increases treatments efficacy but also improves efficiency and economics of beekeeping operations significantly.

[P.11.235] Our bee populations are decreasing in large numbers annually, particularly in areas employing mono-cultures and heavy pesticide usage. The need to dramatically increase bee numbers quickly and effectively

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Brood numbers in a colony require protein consumption, which often occurs through pollination or pollen patties injected with sugar. However, if the lava do not release pheromones, the bees will not consume these large quantities. With a steady slow-release of a carefully pre-determined pheromone dosage, bees will produce more eggs and high-quality royal jelly production increases.

[P.11.236] After 10 years of COLOSS: Who are the COLOSS members and how do they reach out to the beekeepers?

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The mission of COLOSS (Prevention of honeybee COlony LOSSes) is to improve the well-being of honeybees at a global level. To work on the various issues of bee health COLOSS consist of a number of working groups (at present 8) and core projects (at present 3).
The COLOSS B-RAP (Bridging Research and Practice) core project was created in 2014 with the specific intention to deal with knowledge transfer and capacity building in the beekeeping sector. In our 8 workshops so far, we have discussed different aspects regarding extension and dissemination of results from research and practical beekeeping. During those discussions we have realized that it is important to include knowledge regarding human behavioral change, thus social scientists who are working with these topics are members of the group.
Knowing the target group is important for successful knowledge transfer. Generally, the beekeeper is the main target group for COLOSS and consequently for B-RAP. But B-RAP has also a second target group, the COLOSS members. By enabling access to a tool-box available for dissemination and knowledge transfer, we aim to strengthen the overall ability of the organization to bridge the implementation gap. To improve our work with this we need a better understanding of how COLOSS members work with dissemination. Therefore we have conducted a survey among all COLOSS members. Results from the surveys will be presented.

[P.11.237] Distinguishing the components of a bee's nest by semiconductor gas sensors

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The experiment consisted of measuring samples of gas emitted by fragments of variously managed bee honeycombs. The work was performed in laboratory conditions. A multi-sensor detector was used. The instrument, included 6 different semiconductor sensors (by FIGARO): TGS823, TGS826, TGS832, TGS2600, TGS2602, TGS2603.
The research aimed at comparing the response of sensors to gas emitted by various honeycombs: empty ones, with open brood, with sealed healthy brood, with the brood infested by the V. destructor, with storage food, and with bee bread. An empty test chamber was used as an control.
The fragment of each honeycomb was placed in a chamber, and the gas from the tested element was sucked into the multi-sensor

device by means of a pump. The exposure phase of the test sample lasted 10 min., at 21°C. Then there was a 10-min. phase of sensor regeneration by measuring the ambient air.
The sensor readings from the moment of signal stabilization between 101 s and 110 s of the exposure phase were analyzed. Readings of the sensors were within the range of 0.25V-1.68V for an empty chamber, 0.17V-1.81V for the remaining tests and 0.14V-1.52V for the ambient air.
It was shown that each of the sensors has the ability to distinguish the type of brood in the honeycomb. Each of the sensors has the ability to distinguish the open brood from each of the other samples. Two sensors are able to distinguish sealed brood infested with V. destructor mite from each of the other samples. None of the sensors can distinguish the empty honeycomb from the empty test chamber.
Thus, the sensitivity of a multi-sensor measuring device to the gas emitted by bee honeycombs with different contents was demonstrated. This provides grounds for using the device in practical beekeeping to control bee colonies (state and health condition).

[P.11.238] Low power wireless wide area networks for bee colony monitoring and sustainable management

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In this work we investigate and demonstrate low power Internet of things (IoT) solutions for bee colony monitoring. We constructed “smart beehive” equipped with low power consumption sensors, AI and radio communication module in order to perform cloud monitoring system of bee activity & disease control.
To ensure IoT connectivity, reliable bee colony monitoring service and low power consumption, we divide system for basic set of hardware and software components, running with very low power consumption profiles, able to generate and provide primary pre-indicators for swarming probability, disease & pests infestations. And more advanced system – including acoustic analysis - which stays in off mode, and is only triggered by primary system when pre-indicators occur.
Long range, low power IoT connectivity between monitoring hive and Cloud is performed using LORA Technology. System does not send sound samples over IoT, only acoustic signatures. Acoustic signals are re-created on cloud side for further analysis.

[P.11.239] The Energy-Efficient Beehive: A Research Agenda

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Whenever the desired in-hive environment for a honey bee colony’s activities differs from that created naturally by external conditions, the colony must expend energy to establish and maintain the environment it needs. The optimal in-hive environment will depend on whether the colony’s main activity is raising brood, processing nectar, or clustering, while the external environment will vary by climate, season, and weather; and these external factors can vary over timespans from months to hours. While the energy bees expend to maintain their desired in-hive environment is unknown, the likelihood that optimal conditions will occur naturally is low, thus it is possible that bees spend a considerable amount of energy sustaining the in-hive environment, energy that could be used for more productive purposes – if beehives were intentionally designed to minimize bees’ efforts on this task.
The in-hive environmental factors that the colony controls are temperature, humidity, and ventilation. These are interdependent, and bees cannot vary one factor without affecting the others. Likewise, brood rearing and nectar processing often occur simultaneously, as do brood rearing and clustering, and optimal conditions for one of these activities may not be optimal for the other. Thus, defining the optimal in-hive environment may require balancing conflicting needs, while achieving that environment may require a subtle and complex combination of activities.
Optimal conditions are reported to be: for brood rearing: 35C & 60% relative humidity (RH); for nectar processing: low vapor pressure (high temperature and low RH); and for clustering: 7C & 50% RH.
Hypotheses:
1.Well-insulated hives will significantly reduce the energy honey bee colonies expend to maintain an optimal in-hive environment,
2.Providing specific environments for brood rearing, nectar processing, and clustering will enable each to have optimal conditions,
3.Individual and ‘intelligent’ automated ventilation of the brood box during brood rearing, of the honey super(s) during nectar flow, and of the entire hive during clustering will optimize energy efficiency,
4.Colonies in energy-optimal hives will be healthier and more productive than colonies in standard hives.

[P.11.240] Use of voice technologies in apiary records

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It is not easy for beekeepers to take records and collect data digitally during inspection of beehives with the traditional way of data entry. By introducing Speech to Text (S2T) technology beekeepers are able to take digital apiary records and plan their activities more efficient. A strong presence of machine learning and neural computing competence center for S2T and Text 2 Speech (T2S) technologies in Latvia made possible of having voice based hive records. In January of 2017 together with Latvian Beekeepers Association a survey was done

to understand beekeepers' needs and problems concerning apiary records and their use during inspections. We got quite a high response rate with more than 20% of participants willing to apply for early testing of Speech to Text technologies based hive records tool.
In 2018 a first version of voice enabled apiary records system was developed and given to beekeepers for a trial period.
As result of project Beekeepers in Latvia become more active in use of digital hive records and become more responsive with Latvian Beekeepers association using digital tools.
To enable more efficient beekeeping in next development phase voice technologies are being developed towards deeper use of artificial intelligence. For more information about project see here: https://beeking.eu/en/index.html and in recent days it will be published also here: https://enrd.ec.europa.eu/projects-practice_en

[P.11.241] Measurement of temperature in *Apis mellifera* colonies for breeding diagnosis and seasonal development

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The sanitary problems of *Apis mellifera* around the world, lead to a greater surveillance so that beekepers can act rapidly. In order to evaluate the conditions of the breeding or the sanitary conditions, the beekeeper must open the hives, investing time and generating stress. Technology enables to obtain objective information, remotely and in real time. In the bee colonies, some variables which are objectively quantifiable, correlate with aspects which are difficult to qualify subjectively. With information about the temperature of the interior of an *Apis mellifera* colony, a diagnosis could be made and decisions could be taken, faster and effortlessly. One of the ways of obtaining such data, is through a web of sensors. The question that arises is how many sensors should be placed and where. The aim of this research was to take temperature data situated in different parts of a Langstroth hive, and to compare it with the breeding area and its location. Sensors were placed on floors and excluder gratings (9, in 3 rows of 3), breeding chambers (12, 4 on the sides, and 2 at the front and at the back), frames (3, on central positions) and frames with peripheral sensors (7). The gratings were kept above the breeding chambers. Photographs of the 8 hives used, were taken in five opportunities during the spring, and the breeding was measured with the Image J ® programme. The temperature was recorded once an hour using the Arduino software. The results suggest dismissing the use of floors and walls as location for sensors. In both locations, the external factors have a stronger influence than the internal dynamics of the colony. The placement in frames has a high correlation with the presence of breeding, but it makes the handling more difficult for the beekeeper. The superior sensors recorded temperatures that correlate with the measurements performed on the breeding, but they should not be used in excluder gratings by swarming ocurrence at the end of seasons. The research with sensors in central positions and with a different structure should be carried on.

[P.11.242] Use of silage inoculants in the fermentation of ration for *Apis mellifera* bees

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We developed a nutritious, palatable and attractive fermented diet as supplementary food during periods of natural food reduction. For this, two types of commercial inoculants for silages were used: K (only bacteria) and KS (consortium between bacteria and fungi). Thus, these inoculants were used to ferment the protein-based feed, obtaining foods with different fermentation periods: 7, 14 and 28 days. For control, the positive control (C +) - Beebread - and (C-) Negative Control (sucrose solution 50% - w / v) and F (protein base of unfermented ingredients). These feeds were offered, 4 g each, along with sucrose ad libitum solution, for 60 worker bees confined in plastic cages (seven replicates, seven days). A pool of 20 bees / cage was collected on the first day (day 0) and on the 7th day of the experiment, for protein quantification by the Bradford method and for the electrophoretic profile of the proteins by SDS page. We verified that the K and KS feeds of 7 days obtained the best results of consumption when compared with the fresh feed and with the beebread; all the fermented feeds showed higher consumption than the fresh one and beebread. All the bees that ate the fermented feeds (except for the 28-day fermentation period) presented higher titles of protein in the hemolymph when compared to the bees that did not consume any protein food (negative control and day 0).The electrophoretic run presented a protein profile compatible with a good protein expression in the hemolymph of the animals that consumed the fermented feeds, in comparison to the animals without any protein food. Thus, it can be concluded that the fermentation of diets with commercial inoculants for silage has proved to be a viable alternative in the production of nutritive and attractive supplementary foods for *Apis mellifera* bees.

[P.11.243] Impact of polypropylene hives on colony attributes

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Beekeepers around the world are searching for more 'efficient' types of beehives in terms of resistance in time, no need for maintenance, better in thermoregulation, complementary in anti-varroa treatments and they tend to replace the traditional wooden hives with these new

ones made of polypropylene or polystyrene. In an attempt to determine the impact that the polypropylene hives have on several colony attributes, we set up an experiment in Greece and Serbia using three types of hives: a) the traditional wooden hive (wooden brood box and a compact bottom board), b) a combined hive (wooden brood box and a ventilated polypropylene bottom board) and c) the polypropylene hive (polypropylen brood box and a ventilated polypropylene bottom board). For two years we followed several variables such as adult bee population, amount of brood, Nosema prevalence, max and min temperature and humidity on top of the frames, honey production.
The results were similar in the two countries but as expected different between the years. In general, although differences between the three types were not very high in any of the measured variable it was clear that the complete polypropylene hive was able to help in maintaining better thermoregulation as well as lower humidity and it was followed in performance by the combined hive. Same trend was observed in the average amount of brood, population, honey yield and Nosema infestation levels, with the polypropylene hive being always slightly superior followed immediately by the combined type. Although the results cannot be conclusive, it seems that the full polypropylene hive (with its isolation material and ventilated bottom board) can make a difference, especially on the levels of temperature and humidity allowed in the colony. Further tests are needed.

[P.11.244] Challenges for the beekeeping sector when building the future knowledge and innovation system

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In Sweden one sector which experience an increased attention is beekeeping that through its businesses also contribute to pollination in specific areas. It involves many individual beekeepers but is, from a knowledge and innovation system, KIS, perspective, nevertheless highly informal, underfunded and unstructured.
The challenges facing apiculture are many today. One of the most important is related to honeybee health issues, having consequences for rural economy and long-term sustainability. Increased competence and collaboration are seen as central to reach sustainable production systems. But in this respect today's KIS is not functional. Thus, beekeeping illustrates an area that demand social and institutional innovation to be able to deliver public goods as well as sustainable beekeeping businesses.
We need to better understand the key functions enabling environments for responsive multi-actor co-innovation in new areas. The beekeeping sector has good preconditions and might function as a role model for the development of other ecosystem services benefitting the rural economy. If one succeeds in creating a functional and socially robust KIS in apiculture it might be instrumental for the development of KIS for ecosystem services in general.
The implications are that policies need to be not only developed, but also fine-tuned to support
1) the unique characteristics of networks and platforms for learning and co-innovation in new areas of development, 2) systems for vertical integration of actors in the policy chain and 3) action evaluation for continuously improving pre-conditions and methods when scaling up and out social and institutional innovations.
Relevant stakeholders are identified and participating in workshops as well as interviews. A qualitative analysis is made based on collected data as well as earlier research on KIS. The qualitative analysis is guided by triangulation of the different data input, including a Delphi-inspired methodology.
The results include a) recommendations on how a socially robust KIS for the Swedish beekeeping sector might be organised and function, b) a critical analysis of the main challenges when building a KIS from scratch, and c) consequences for other, contemporary areas such as the development of new KIS for existing ecosystem services.

[P.11.245] Exploration and research on modern beekeeping in China

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China is a large beekeeping country, but not a powerful nation on apiculture. The contradiction between the development of crop pollination, the booming domestic demand for bees, the improvement of honey quality and the shrinking of China's bee industry, such as backward production mode, industrial organization and low degree of mechanization, requires the transformation and upgrading of the apiculture of China.
Modernization and large-scale beekeeping is the only way for China's apiculture. At present, there already has mature modern apiary in Xinjiang of China. Based on the technological innovation of bee rearing, tray beehives, shallow super box and other proprietary intellectual property rights, the apiary has realized the mechanization of the whole process of beekeeping, bee breeding, colony transfer, honey harvesting, purification and filling.
In addition, the package bees technology invented by China can maintain the healthy bee colony for more than 10 days in the long transportation, which can provide reference for Canada, the United States and other countries in the problem of bees overwintering.

[P.11.246] NanoBeekeeping: Applications of nanotechnology in beekeeping

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Nanotechnology depends on using nanoscale materials. Recently, nanotechnology has been applied in many agricultural aspects but still few studies have been done on beekeeping. This study presents potential applications of nanotechnology in beekeeping which can lead to open new trends for beekeeping development. Basically, nanotechnology can be applied on many aspects in beekeeping including; beekeeping tools, productivity aspects, control of bee pest and diseases, increasing plants pollination and bee products. Implementing nanotechnology with beekeeping tools can help boosting the ability of bees to survive under harsh environmental conditions and to produce more hygienic tools, nanoparticles can help improving the control of bee pests and diseases, some nanoparticles can be helpful to boost plant pollination, bee feeding can be boosted when nanotechnology is used, and bee products prepared as nanoparticles have many medicinal and nutritional properties which can increase marketing value. There are many techniques that can be used to produce nanoparticles. Therefore, the potential applications of nanotechnology in beekeeping need great research efforts to find out the best formulation to accomplish each task. Nanotechnology can be also used in line with other techniques including geographical information system to improve honey bee health. This study presents new trends and encourages researchers to consider nanotechnology during their studies. Additionally, the potential hazards of nanoparticles in the agro-ecosystem are presented.

[P.11.247] Subspecies variability of *Apis mellifera* in the coastal zone in O'Higgins region of Chile

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The O'Higgins Region in Chile, is the region with the highest national beekeeping activity because it has the largest number of colonies with high standards in pollination services, production of honey and biological material (for national and international markets such as Canada and France). Until a few years ago in the region, the predominant genetic lineage was *Apis mellifera ligustica* because it was the most appropriate according to the ecology of the area. Today, it has been seen that *Apis mellifera* carnica predominates but it is not known how relevant its population or its distribution in the region. The degree of purity or hybridization of the lineage is also unknown. On the other hand, it is questionable whether this change in breeding carnica lineage has been a good or bad decision, as it seems less suitable for the conditions of temperature, humidity and nectar flow, and maybe is a factor that influences the high mortalities and low production in the central area. It is because of this, a study was carried out to evaluate the genetic diversity of honeybee colonies in the coastal area of the O'Higgins Region using Geometric Morphometry using the Morpho J® program. This method allows to calculate the similarity in wing of the shape of samples obtained with reference wings of 6 subspecies of *Apis mellifera* across the Mahalanobis distance. Five apiaries were sampled from each of the 5 communes present in the coastal zone with a total of 3 colonies per apiary, obtaining from them 30 workers to obtain 15 wings per colony. The results indicated that for the area studied the predominant subspecies was *Apis mellifera* carnica. The above indicated that the genetic pattern for the central zone of Chile could have changed in recent years, it is also proposed to introduce the appropriate genetics, revert the productive purpose of these apiaries to the production of biological material, since it is considered that the ecological conditions of the region is favorable for queen breaders, which should be complemented with a study of the pool of lineages present in the region.

[P.11.248] Production and use of fresh apicultural pollen in supplementary feeding of africanized bee colonies

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In Brazil, during part of the year there is a great availability of food resources for bees, but there are periods where there is a restriction or even total absence of food. In Northeast Brazil, this situation can last up to eight months. In the absence of food in the wild, there is a need for additional feeding to the swarms, to avoid the reduction of the populations and the abandonment of the hives. Supplementary feeding in certified organic beekeeping requires natural and non-GM food. Storing and using surplus pollen and honey can be a supplementation alternative for the shortage period. This work quantified the pollen that could be collected in the flowering period and used this pollen in the supplementary feed (sugar + honey + pollen) as an alternative for the maintenance of the swarms. In the period from April to June 2017, in Teresina / PI, pollen was collected from nine hives and later the fresh pollen was homogenized and mixed with sugar and honey to feed hives during the period of food shortage. During the collection period a total of 10.8 kg of pollen was obtained for the 9 hives, an average of 1.2 kg of pollen/hive. The mean crude protein content of the homogenized pollen was 31.88%. In the feeding of the hives during the supplementation period the average consumption of the mixture was 52 g/hive/day. Thus, pollen collected in 60 days in a colony can be fed for 145 days, using a mixture with a proportion of 16% of pollen added to sugar and honey. The results show that the collection of pollen in the flowering and its use as a source of protein in a supplementary feed in the period of scarcity allows the maintenance of the swarms, since there was no loss of swarms in the hives fed.

[P.11.249] Apiter laboratories - apicultural technology and quality

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Apiter Laboratories is a pioneer in the development of technology and processing of Propolis in Latin America, it is a Uruguayan company established in 1975.
The main purpose of the company is the processing and commercialization of propolis extracts and pharmaceutical products, with a strong development in research and technological activities related to the harvest, processing and formulations of Propolis for therapeutic purposes.
APITER has implemented a Quality Management System in accordance with the ISO 9001/2015 Standard applicable to the design, development, production and commercialization of Propolis extracts.
This implementation consolidates the continuous improvement of the system, the traceability of the Propolis extract to its origin and the obtaining of a database of evaluated and qualified producers that make up the Quality Management program and allows the continuous improvement in the sanitary management of Beehive.
Uruguayan Propolis comes from a privileged geographical region called Sabana Uruguayense, with botanical species defined as Eucalyptus globulus, Populus sp., Betula sp., And Salix sp., among others that originate a proplis extract of superior quality in active principles.
Apiter has implemented an innovative Propolis harvesting system through meshs trays, obtaining a fresh Propolis with higher Polyphenols content and a lower level of contamination.
In 35 years of research, technological development and implementation of traceability, the production process has been optimized, from which standardized Propolis extracts are obtained, which ensure their composition, stability of their active ingredients and the reduction and/or elimination of contaminants.
The Propolis extract of Apiter contains a high percentage of polyphenols and flavonoids; determining that it is one of the highest in the world. Flavonoid levels range from 135-250 mg/g of PEP-100 expressed in galangin.

[P.11.250] Apenova

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APENOVA is a low-cost, easy-to-use operating procedure that can redevelop and enhance beekeeping at local and national level. APENOVA is based on the technique of the division of bee families and the use of real cells. It allows to implement an effective fight against varroa, to increase the number of hives every year, to limit the use of acaricides but above all to promote the spread of the breed of bees that best suits a territory. Bees that are selected for the qualities DO.PA.MI.NOS. (docility, resistance to diseases, excellent production of honey, poor propensity to swarm). The APENOVA project involves the collaboration of beekeepers, producers of real cells, bees technicians and research institutes.

[P.11.251] Gastric digestion comparisons of bee pollen with or without wall-disruption in vitro or in vivo

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Bee pollen collected by honey bees contains a substantial amount of nutrients and has a high nutritive value. However, a high level of nutrients can be difficult to be digested and absorbed due to the complex walls of bee pollen. Our results reveal that amino acids are mostly distributed inside the pollen wall using TOF-SIMS (Time-of-flight secondary ion mass spectrometry). To crush pollen walls, lotus bee pollen, rape bee pollen, apricot bee pollen, wuweizi bee pollen and camellia bee pollen were treated with a combination of ultrasonication and high shear technique (US-HS). After treatment, the pollen wall is entirely broken into fragments and the content of almost all nutrients increase. We investigated the effects of saliva, gastric and intestinal digestion on bee pollen with or without wall-disruption in vitro and in vivo. Our results reveal that bee pollen grains without wall-disruption cannot be digested by the gastrointestinal tract, they will be integrally excreted using fluorescence trace technique. Thus, wall-disruption seems to be a feasible method for improving pollen nutritional digestibility and absorption.

[P.11.252] New propolis type from Changbai Mountains in north-east China: chemical composition, botanical origin and biological activity

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Propolis is a bee product with a wide range of biological activities and its chemical compounds depend highly on the type of plant accessible to the bees. Changbai mountains are a major mountain range in Northeast China and are one of the major producing areas of bee products in China. In this study, we evaluated the total phenolic acids and flavonoid contents as well as the antioxidant activity of

propolis sampled from the Changbai mountains area (CBM). We also identified major compounds and qualified their contents by HPLC-ESI/MS and HPLC-UV, and found that the content of p-coumaric acid and an unknown peak (CBE) in CBM propolis was higher than propolis from other parts of China. The unknown compound CBE was isolated, purified, and identified as benzyl p-coumarate by MS and NMR. The plant sources of CBM propolis are Populus davidiana dode and Populus simonii Carr, which widely distribute in Changbai mountains area. Furthermore, we evaluated the anti-proliferation effect of CMB propolis on different tumor cell lines, discovered that CMB propolis was able to inhibit the proliferation of human melanoma cell line A375 and gastric cell line SGC-7901 significantly.

[P.11.253] Sensory descriptive profiling of Flow® Frame honey compared to honey extracted using conventional methods

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During conventional processing, honey undergoes centrifugation, heating and filtration. Such processing may lead to changes in the chemical profile resulting from loss, degradation or generation of secondary reaction products. Consequently, the method of processing may also impact honey flavour and sensory perception.
In 2015 a novel method of honey extraction was introduced in the form of the Flow® Hive. This new technology enables honey extraction directly from the hive. Specially designed honeycomb frames replace the static wax or plastic foundation frames of a conventional beehive in the honey super. When the Flow Frames are split during harvesting honey flows down through a central axis and out via a channel to the rear of the hive where it is collected directly. This technology eliminates the need for hot-knife decapping, centrifugation and even filtration of the honey. The aim of this study was to determine if the honey extracted from Flow Frames would have different sensory properties and volatile flavour profile compared to honey extracted by conventional methods.
Descriptive sensory profiling was conducted on individual honeys produced from two Australian native floral sources - Macadamia sp. or Pultenaea spp. (yellow pea) - and three honey extraction treatments - Commercial, small-scale conventional, and Flow Frame. Sensory profiles obtained by 10 panellists showed that the honey samples produced using the Flow Frame method had higher scores for floral aroma in both floral sources, and had higher herbaceous aroma in the yellow pea samples and higher scores for citrus aroma in the macadamia samples. The Flow frame honey also received lower scores for pungency and lingering aftertaste. This study showed that the Flow Frame honey production method has a positive impact on honey sensory characteristics independent of the floral source.

[P.11.254] Biogenic Synthesis of Silver Nanoparticles (AgNPs) using Propolis Extract, their Characterization, and Biological Activities

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Nanoparticles fabricated through plant extracts are getting attention in the field of biology because of their biocompatibility, green approach, and eco-friendly nature. The present study reports the biogenic synthesis of silver nanoparticle (AgNPs) using propolis ethanol extract (PeExt), their effects on immune modulation, antibacterial, and hemolytic activities. The characterization of NPs fabricated with extract (PeExt-AgNPs) was performed by ultraviolet-visible (UV-Vis) spectrophotometry, scanning electron microscopy (SEM), and Fourier-Transform Infrared Radiation (FTIR) spectroscopy. Both PeExt and PeExt-AgNPs revealed significant effects ($p < 0.05$) against Gram-negative (Escherichia coli, Proteus mirabilis, Shigella flexneri) and Gram-positive (Staphylococcus aureus) bacterial strains. The results also revealed that there were low stimulatory effects of PeExt on normal and phytohaemagglutinin (PHA) stimulated rat splenic cells. The stimulatory effect decreased with the decrease in the concentration of PeExt. PeExt and PeExt-AgNPs were safe to the vital organ (liver) and red blood cells (RBCs). They didn't cause any severe elevation in liver enzymes.

[P.11.255] A Comparative Study on the Two Different Methods IRMS and CRDS for Estimation of ™ 13C (‰) of Honey Samples

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We have analyzed the isotopic ratio of carbon (™13C ‰) of honey samples (n=10) employing two different methods namely IRMS and CRDS. In general, the honey samples satisfied the specification suggested by codex. The ™ 13C values for the tested honey samples ranged from -12.0 to -26.6‰. A lower value indicates the possible adulteration of honey with sugar syrup or honey from the beehive in which bees were fed sugar syrup coming from C4 plants. Results obtained from two methods showed that there is no significant difference in the values for respective honey samples. However, higher accuracy was observed in the results obtained from CRDS system.

Comparable and consistent results, lower maintenance cost and ease of handling make CRDS system a better alternative to traditional state-of-the-art IRMS method for carrying out routine analysis of honey samples.

[P.11.256] Antioxidant Activity of Different Types of Honey Produced by Different Methods

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Many processes affect the chemical and physical properties of honey. Antioxidant activities and physico-chemical characteristics were studied. Two types of honey: citrus honey and onion honeys were used in the current study. Ripe and unripe honey from the comp was directly taken without extraction or filtration, or any other processing procedures in order to avoid changes or to prevent any of its components. This study reports the physico-chemical characterization and antioxidant activities of different types of citrus and onion honey. Fresh honey lyophilizes honey and honey stored for one and two years were tested. The DPPH method was used to assess the antioxidant efficacy. Results reported that total phenolic content of citrus honey was higher than onion honey with 0.95 and 0.79 mg/100 g, respectively. Also, the antioxidant activity was higher in citrus honey than in onion honey. Considering the results, characteristics of honey samples are highly dependent on the floral origin, processing procedures and storage.

[P.11.257] Determination of the effect of different extraction solvents and methods on phenolic profile, total phenolic content and antimicrobial activity of propolis

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Propolis is a complex material of resinous substances which is collected by bees from the exudates of plants and modified by mixing it with enzymes, pollen and wax. Propolis of which chemical composition mainly depends on the botanical and geographic origin, has high level of phenolic and flavonoid content.

The objective of this study was to compare the effect of different extraction methods and solvents on the total phenolic content, phenolic profile and antimicrobial activity of propolis. Propolis samples collected from five different countries (Argentina, Bulgaria, Brazil, Iran and Turkey) were extracted by using two different extraction methods (ultrasonic and shaking) with three different solvents (propylene glycol, %70 and %96 ethanol). Propolis samples were extracted for three times to reach the maximum yield of extraction. The obtained extracts were analyzed for phenolic content by HPLC PDA. With this method, propolis samples were screened for 38 phenolic compounds and 18 of them were absent for all samples. Total phenolic content of propolis samples were determined by UV Spectrometer and expressed as gallic acid equivalent. The antibacterial activities of propolis samples against S aureus and E.Coli were determined by micro dilution method. Results were expressed as Minimum inhibitory concentration (MIC) in mg/l and minimum bactericidal concentration (MBC) in mg/l.

There was no significant difference in terms of the total phenolic content (TPC) and phenolic profiles of samples extracted by ultrasonic and shaking method. In terms of the total phenolic content, different solvents showed no significant difference for the samples collected from Iran, Argentina and Turkey. However, 70% ethanol was found more effective solvent for the samples collected from Bulgaria and Brazil. In view of phenolic profiles extraction with hydroalcoholic solutions (96% and 70% ethanol) were found more effective than propylene glycol solution. There was no significant difference between extraction methods in terms of antimicrobial activity but hydroalcoholic solutions showed higher antimicrobial activity than propylene glycol solution.

[P.11.258] Colombian bee pollen: phenolic and flavonoids antioxidant potential free radical scavenging and physicochemical properties

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Corbicular bee pollen is a natural product collected by bees that after a delicate process of agglomeration, and stabilization, by drying, has gained commercial importance in the marketing of natural products due to its nutritional and therapeutic properties. The goal of this work was focused to study of total flavonoids and phenols, antioxidant potential and radical scavenging activity of corbicular bee pollen ethanoic soluble compounds. Antioxidant capacity was measured by FRAP and ABTS for radical activity. flavonoids were identified by HPLC. Phenolic fraction observed varies between (11.8 ± 2.22 and 30.6 ± 7.10) mg EAG/g, flavonoids between 5.67 ± 2.35 and 12.1 ± 1.13 mg Eq. Quercetin/g pollen. The reducing capacity (11.8 ± 2.22 to 30.6 ± 7.10) and Trolox/g (9.00 ± 3.43 and 17.9 ± 4.30) µM for FRAP and ABTS respectively. Pigments and soluble compounds present in the hydroalcoholic matrix, exhibit yellow and orange-yellow tones, revealing the presence of carotenes, xanthophylls, phenolic compounds between acids and flavonoids (chlorogenic, gallic, ferulic, p-coumaric and caffeic acid as well as Isoquercetin, myricetin, herbacetin, quercetin, luteolin, kaempferol, and isorhamnetin. The phenolic compounds associated with bee pollen, are metabolites such as apigenin-7-O-glucoside, luteolin-7-O-glucoside, quercetin-3-glucoside, isoflavones, glucosides of genistein, dihidroflavonoids, of Quercetin, Isorhamnetin-3-O-glucoside, and Isorhamnetin-3-O-glucoside, derived from cinnamic acids. The results observed are relevant in terms of nutritional supplement with Nutraceuticals properties under the functionality of its components.

[P.11.259] Comparison of chemical composition of floral honey and sugar fed honey made by honey bees (*Apis mellifera*)

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In this study, we investigated and compared some chemical properties of Korean natural honey and sugar-fed honey for their quality characteristics. The natural honey samples were monofloral from chestnut and acacia flowers, and the sugar-fed honey samples were collected from honey bees feeding on sugar cane and sugar beet. The chemical properties of the honey samples, such as moisture, total proteins, total lipids, ash, carbohydrate, minerals, vitamins, and free amino acids were determined. The moisture content was 18.5±0.9% in natural acacia honey, 17.2±0.9% in natural chestnut honey, 19.6±0.9% in sugar cane-fed honey, and 24.8±2.9% in sugar beet-fed honey. The total protein and ash contents were the highest in natural chestnut honey. Maltose and sucrose were not detected in natural honey but were detected at 2-7% in sugar-fed honey samples. The vitamin, mineral, and free amino acids contents of natural honey were higher than sugar-fed honey. The natural chestnut honey is the highest in honey samples. These results confirmed that the quality of natural honey was better than that of sugar-fed honey. Also, the vitamin, mineral, and free amino acids contents are potential characteristics for distinguishing between natural and sugar-fed honey.

[P.11.260] Procedure of honey extraction influencing the pollen composition during melissopalynological studies

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The purpose of this study was to investigate the suitability of honey extraction methods for palynological analysis to determine botanical origin of honey. We collected the honey samples via three modes viz. extracted honey, squeezed honey and sucked honey. Here we considered two native honey bees (*Apis dorsata* and *Apis florea*) and one introduced honey bee (*Apis mellifera*). The number of pollen grains in extracted honey and squeezed honey was significantly higher than sucked honey. Which indicates that some pollen grains in extracted honey and squeezed honey were came from stored pollen loads or bee bread in the hive. Hence, pollen spectrum of sucked honey is more accurately denotes the source of bee foraging nectariferous plants. Among the analyzed honey samples, most were multifloral in botanical origin and only few were unifloral type. Those were obtained during late autumn (Eucalyptus globulus type), winter (Brassica spp. type, Eucalyptus globulus type), spring (Borassus flabellifer type, Butea monosperma type and Lannea coromandelica type) and summer (Sesamum indicum type, Terminalia arjuna type). Predominant pollen type was lacking in monsoon and autumn and manifested dearth of resources for all the honey bees. The bee pasturage for these three honey bees in West Bengal was almost similar. They utilized large number of wild as well as cultivated plants. All the honey bees preferred tree habit with small sized flowers as a source of nectar. The most common shapes of the bee visited flowers were dish-like and brush-like, and the preferred flower colours were yellow, white or cream.

[P.11.261] Determination of proline content in different parts of comb honey

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Turkey is an important beekeeping country due to its climate, flora and biodiversity. Especially, different climatic conditions of Turkey own are creating a suitable environment for migratory beekeeping. To make best use of the flora, bee colonies are moved between various regions according to different seasonal conditions and the need of bees in migratory beekeeping.

The content of honey may vary depending on the climate, flora, seasonal conditions, colony need, pollen/ nectar source and production technique of beekeeper. Since migratory beekeepers follow the flora, they visit various regions without harvesting honey. This situation sometimes causes difficulties to establish standardization and common quality criteria. Especially migratory beekeepers, produce comb honey from different regions, have suffered some difficulties in meeting the proline values stated in standards and it is requested to do research on this subject.

In this study, comb honey samples were collected from beekeepers producing honey in migratory beekeeping conditions. It was investigated whether there are any differences between the utilization levels of honey bees from these plants, proline contents and prolin quantities at different parts on the same comb honey.

[P.11.262] Characterization of the Apitoxin from *Apis mellifera syriaca*, a honeybee from the Middle East region

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Apitoxin is a mixture of several components, with proved therapeutic benefits, among which anti-inflammatory, pain killer and various

cardiovascular conditions. In this work, we extracted Apitoxins from *Apis mellifera syriaca* honeybee at different seasons and checked their respective activities. The proteomic content and biological properties of the crude apitoxin was analyzed. Using high-performance liquid chromatography-tandem mass spectrometry, we evidenced the apitoxin main molecules, such as phospholipase A2, hyaluronidase, adolapin, apamin and melittin. More in depth test were applied to some molecules and for biological activities. Alongside, hemolytic activity was observed in human blood subjected to the apitoxin at high doses. Apitoxin and its derived melittin displayed antioxidant activities and not surprisingly pro-inflammatory effect. Anticancer tests were also applied. Positive results would open new avenues for novel drug design.

[P.11.263] Evaluation of Antioxidant Activity, Phenolic, Flavonoid and Vitamin C Contents of Several Honeys Produced by the Indonesian Stingless Bee: *Tetragonula laeviceps*

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Indonesia is an archipelagic country with different geographical regions. The different geographical for the meliponiculture, species of stingless bees and the types of plants that serve as food sources influence honey quality (chemical composition). The objective of this study was to an evaluation of antioxidant activity, total phenolic content (TPC), total flavonoid content (TFC) and vitamin C content of honey produced by the Indonesian stingless bee: Tetragonula laeviceps from different geographical regions. Honey was obtained from three regions, the Faculty of Animal Science Universitas Gadjah Mada (UGM), Nglipar Gunungkidul, Yogyakarta and Klaten, Central Java. The TPC and TFC were determined by the spectrophotometric method, antioxidant activity by DPPH assay method, vitamin C by the 2,6-dicholorophenolindophenol titrimetric method. All data were statistically analyzed using one-way analysis of variance (ANOVA) followed by honestly significant difference (HSD) test. The results showed that the geographical region had a highly significant effect ($P<0.01$) on TFC, and antioxidant activity. In addition, the geographical region had a significant effect ($P<0.05$) on TPC but not on vitamin C ($P>0.05$). The highest antioxidant activity was in honey from Klaten, at 91.23±0.41%, followed by honey from the Faculty of Animal Science UGM 90.49±0.86%, and the lowest was in honey from Nglipar 47.32±0.46%. The highest TFC was in honey from the Faculty of Animal Science UGM 902.10±121.92 µg/g QE, followed by honey from Klaten 764.22±23.33 µg/g QE, and the lowest was in honey from Nglipar 212.41±44.78 µg/g QE. The highest TPC was in honey from the Faculty of Animal Science UGM 1.69±0.33 %w/w GAE, followed by honey from Klaten 1.21±0.46 %w/w GAE, and the lowest was in honey from Nglipar 0.54±0.08 %w/w GAE. Vitamin C content of honey from the Faculty of Animal Science UGM, Klaten, and Nglipar were 7.88±1.76, 6.51±0.71, and 5.67±0.38 mg/100 g, respectively, was similar. It can be concluded that honey from the Faculty of Animal Science UGM and Klaten have the highest TFC, TPC, and antioxidant activity compared with the honey from Nglipar.

[P.11.265] Physicochemical parameters of liquid and spray-dried honey of *Scaptotrigona pectoralis*

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Certain physical and chemical parameters 24 of honey [liquid and powder (spray-dried)] produced by the stingless bee *Scaptotrigona pectoralis* from Yucatan, Mexico, were determined. Measured parameters in liquid honey were as follows: color (85.0 mm Pfund), moisture (25%), pH (3.6), acidity (116.0 meq kg-1), ash (0.25%), hydroxymethylfurfural (43.0 mg kg-1), invertase activity (22.0 IN) and reducing sugars [38.0 g (10 kg-1)]. Of nineteen volatiles compounds positively identified, p-anisaldehyde was found as the major compound. The antioxidant activity was evaluated by using the methods ABTS•+ [37.0 µmol ET (10 kg-1)] and DPPH [3.50 µmol ET 10 kg-1]. Polyphenols content was 23.0 mg EAG 10 kg-1. Honey antioxidant activities were by ABTS•+ [37.0 µmol ET (10 kg-1)] and DPPH [3.5 µmol ET (10 kg-1)]. Polyphenols content was 23.0 mg EAG (10 kg-1). On the other hand, the spray-dried honey of S. pectoralis, presented high hygroscopicity [22.0 g (10 kg-1)], good solubility (265 s), low density (0.34 g ml-1) and good volatiles retention (77%). The antioxidant activity of the honey powder was higher [51 µmol ET (10 kg-1)] (ABTS•+ method) than in the liquid honey. The results showed new information about the composition of the liquid honey and honey powder, which would help for assessing their qualities. Further, spray-drying process exhibited as alternative to preserve and even improve the antioxidant activity of S. pectoralis honey due probably to enrichment of antioxidants compounds in the powder.

[P.11.266] A comparative study on the biochemical, antioxidative and anti-microbial activity of *Apis* and *Trigona* honey collected from different geographical areas of India

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Honey is one of the very well-known bee products which have been exploited medicinally from ancient times. It has been used for enhancing the human health in many possible ways due to their nutritional and therapeutic values. Honey is used traditionally for treating burns and wounds which are pathogenic infected. It is a proven natural energy giving food and an excellent immunity builder. It is packed with anti-oxidants and flavonoids which help reducing cancers and heart ailments. The natural Apis honey and *Trigona* honey samples collected from different geographical areas were selected and anti-oxidative activities were tested and compared with processed honey and with other bee products. Hydroxymethylfurfural (HMF) content was tested in honey samples for analyzing their stability. Protein contents in honey samples were estimated by Lowry's method and proline content was analyzed in each sample. Their antagonistic property against two Gram positive and two Gram negative using agar well diffusion method.

[P.11.267] Effect of two different solvents on antibacterial activities and isolation bioactive of Iranian propolis compounds

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Considering the importance of propolis as a natural product for their remedial and therapeutic value, bioactive compounds of two different solvents of four Iranian propolis extract were compared by GC-MS analysis. Then their antibacterial activities were evaluated against Escherichia coli, and Staphylococcus aureus using disk diffusion antimicrobial method. Comparison between the two utilized extractions indicated that the total amount of flavonoids in Dichloromethane extracts of all four propolis regions was higher than their ethanolic extracts. Pinostrobin chalcone identified in both Ethanolic and Dichloromethane extracts of the same propolis samples of Hamedan province increased from 0.31% to 3.95%. The antimicrobial activities of Ethanolic and Dichloromethane extracts of propolis against E. coli and S. aurous confirm this assumption. Results indicated that solvents are important for extracting flavonoids, phenolic acids, and terpenes as main active components of propolis.

[P.11.268] Near-Infrared analysis applied to Morocco honeybee (*Apis mellifera intermissa*) venom: A first approach

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Honey bee venom, Apitoxin, exhibits different pharmacological activities, among them anti-inflammatory, antibacterial, antimutagenic, radioprotective, antinociceptive and anticancer activity. The quality assessment of this bee product is commonly achieved by chromatographic techniques, which enables the identification of the major protein and peptides. In this work we used the infrared spectroscopy to explore the differences between fifteen samples of bee venom collected in three regions of Morocco (Northeast, Center, and South of Morocco). Near-Infrared spectroscopy (NIR) was employed, and the spectra were recorded using a BRUKER spectrometer operated with the OPUS®, version: 7.5.18 (Bruker Optik, Germany) software in reflectance mode. About 10 mg of bee venom was poured into a clean glass vail with 22 mm diameter. The final spectra, collected with a resolution of 8 cm-1, was a result of 32 scans in the wavenumber range between 4000 and 10000 cm-1. According to the spectra, there is a noticeable IR information in the regions from 9000 to 4500 cm-1. The absorption bands for polypeptides and proteins (the main compounds of the dry honey bee venom) appears between 4800 cm-1 and 6800 cm-1 in the overtone region. However, this product has also low molecular weight compounds that add relevant spectral information to be discussed. In the more relevant spectral region, principal component analyses (PCA) were performed using the Unscrambler® X version: 10.5.46461.632 (CAMO Software AS, Oslo, Norway). Concerning the majority constituents, such as (Melittin or Phospholipase A2), the samples revealed differences. Additionally, high sensitivity to the storage period of samples, was revealed by this method, which allowed the discrimination among old and fresh samples. In conclusion, from the preliminary data collected with this matrix, NIR shows important potential to be a powerful tool for its quality control
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[P.11.269] Melittin identification in bee venom and antimicrobial activity evaluation against Staphylococcus aureus

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Melittin is the main component of bee venom comprising 26 amino acid residues with amphipathic characteristics. These residues allow melittin to interact with lipid membranes and to increase the permeability of cell membranes triggering its lysis, mainly through pore formation. Melittin is recognized due to its antimicrobial activity and is studied as an alternative for treating infections caused by drug-resistant microorganisms. In this work, we achieved the purification of the peptide from *A. mellifera* venom using fast protein liquid chromatography (FPLC), and we confirmed its identity by nanoLC MS/MS. The purification method allowed us to remove all detectable contaminants from melittin such as the bee venom allergen phospholipase A2 (PLA2) as well as other compounds with molecular weight similar to melittin like the smallest neurotoxin apamin (2 kDa). The purification process let us to obtain an active peptide with a calculated molecular weight of 2.8 kDa and antimicrobial action against a multiresistant strain of the Gram-positive bacteria Staphylococcus aureus. The antibacterial action of bee venom was also assessed. The minimum inhibitory and bactericidal concentration were respectively: bee venom (113 and 225 µg ml-1) and melittin (104 and 207 µg ml-1). Future studies using combinations of bee venom compounds like PLA2 with melittin are needed to evaluate a reduction in MIC and MBC values product of a possible synergistic effect between melittin and the former compound that could enhance the antimicrobial effect against S. aureus and other Gram positive and Gram negative bacteria.

[P.11.270] Fingerprint NMR based on the chemical profiling of colombian red propolis

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Propolis is a complex material of resinous consistency produced by bees which has a highly variable physical appearance, color, and consistency, depending on many factors such as geographic origin, types of vegetable sources, time of collection, and season of the year. Chemical composition is extremely complex and variable. Tropical propolis has chemistry quite different from that of temperate-zone propolis. Moreover, its composition is highly variable because of the richness and versatility of tropical flora. In order to progress towards a method for the quality control of this complex material, 1H-NMR approaches as methods of quality control have been compared. Despite the diversity of Colombian propolis, never before been studies on red propolis. The present study aimed to investigate the chemical profile, from an insular zone of San Andres, Colombia. Seven samples of raw red propolis were carried out by 1-HNMR spectroscopy (0.2 g sample/1 mL methanol deuterated/TMS/400 MHz). Chemometric analysis indicated differential fingerprint show distinct chemical profile respect of others samples reported in scientific literature. Rutin, liquiritigenin, daidzein, formononetin, quercetin, biochanin A and gutifferone E, vestitol and neovestitol, and medicarpin are principal chemical markers.

[P.11.271] Interactions between honeys and yeasts for mead production

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Mead is a traditional alcoholic beverage, containing from 8 to 18% of ethanol, resulting from the fermentation of dilute honey solutions by yeasts. Its production still remains a relatively empirical process and several problems are usually encountered, namely delayed (lasting several months to years) or stuck fermentations, unsatisfactory quality parameters, lack of uniformity of the final products and unpleasant sensory properties. The fermentation rate depends on several factors, such as honey variety, yeast strain, yeast nutrition and must pH.
The aim of this work is to optimize the mead fermentation through honey-must supplementation with available nitrogen, salts and/or vitamins and to evaluate the synergies between honeys and yeasts. For this study, musts, obtained from four different honeys (Orange blossom and sainfoin blossom honeys from Italy and two multiflora honeys from Belgium) were fermented by various active dry yeast strains. The fermentation kinetics were monitored and the physicochemical characteristics of final meads were evaluated i.e. ethanol concentration, density, organic acid and residual sugar concentrations, chromatic characteristic, flavour profile (esters and higher alcohols). Our results indicate that the fermentation profiles were greatly influenced by the type of honey and, to a lesser extent, by the yeast strain. Moreover, for single flower honeys, specific nutrients, such as available nitrogen, minerals and/or vitamins, are required to obtain fast fermentation and high ethanol levels. Sensory analysis of the final meads highlighted the impact of yeasts on their final bouquet and revealed adequate honey-yeast partners for mead production.

[P.11.272] Sensory training to analysis quality of honey from hives feeded with supplementary feeding

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Sensory analysis of bee honey is an important tool for determining its floral origin, for subsequent quality control practices and the consumer preferences towards this product. The aim of this research was the development of a protocol for training and monitoring of assessors. Descriptive quantitative analysis was conducted to evaluate the sensory properties of honeys produced by honeybees feeded with different sugars pastes: sugar paste (Apipasta®), Sugar paste with vitamins and free aminoacids (Apipsta Vitamina®) and sugar paste with vitaminas and crude protein (3%) (Apipasta Plus®), all manufacturated in Zukán S.L (Murcia, Spain) in comparison with control group (non feeded).
Samples of honey from four groups were evaluated by a panel composed with 8 assessors. The following descriptors have been evaluated: sweet, acidity, bitterness, salty, as well 8 families of odour, flavor descriptors and appearance descriptors. The evaluation of each descriptor was performed on a linear intensity unstructured scale of 10 cm. The panel was monitored to evaluate the efficiency of the training protocol developed.
The percentage coincidence test indicated that the training protocol developed was effective and the panel was prepared to evaluate the honeys samples. No significant statistical differences in odour, flavour and basic taste were found in all analyzed honeys ($P > 0.05$) independently of the type of feeding. For the visual attributes, only differences were found for the color. Control honey and honey from hives feeded with Apipasta® had a relatively higher value than honeys from feeding bees with Apipasta Vitamina® and Apipasta Plus® although it was not relevant in quality of honey.

[P.11.273] Development of nanoemulsion containing royal jelly and melittin from *Apis dorsata* bee venom with anti-hyaluronidase activity

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Royal jelly and bee venom are products from honey bees which are widely used in food and cosmetic industries. The aims of the present study were to investigate the anti-hyaluronidase activity and irritation properties of royal jelly and bee venom from *Apis dorsata*. Additionally, nanoemulsions containing royal jelly and bee venom were developed for anti-hyaluronidase purpose. Royal jelly and melittin from bee venom of *A. dorsata* were investigated for anti-hyaluronidase activity by in vitro spectrophotometric method. Moreover, the irritation properties of these bee products were investigated using hen's egg test on the chorioallantoic membrane assay. The bee products with anti-hyaluronidase activity with no irritation effect was selected for nanoemulsion development. Nanoemulsions were developed using sonication method. Various factors affecting the nanoemulsion formulations, including types of oil, types of surfactant, amount of oil, and amount of surfactant were investigated. The nanoemulsions were then characterized for particle size, polydispersity index (PDI), zeta potential, and stability in heating-cooling condition. The results noted that royal jelly and melittin possessed potent anti-hyaluronidase activity with the inhibition of 91.4±12.2% and 95.0±8.7%, respectively, which were higher than that of epigallocatechin-3-gallate (90.94±4.86%). Therefore, royal jelly and melittin were used in the nanoemulsion development. It was noted that types of oil, types of surfactant, amount of oil, and amount of surfactant affected the nanoemulsion characteristics. The most suitable nanoemulsion, contained 10%w/w rice germ oil, 15% Tween 20, 75% deionized water. The nanoemulsion containing royal jelly and melittin were stable after the stability test with the size of 314.80±4.04 nm, PDI of 0.26±0.04 and zeta potential of -0.04±0.08 mV. Therefore, the nanoemulsion containing royal jelly and melittin form *A. dorsata* would be an attractive anti-hyaluronidase formulation for further used in cosmetic industry as anti-ageing product.

[P.11.274] Characterization and differentiation of colombian honey through application of sensory instrumental tools and physicochemical properties

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In spite of the immense natural richness existing in Colombia and its great biodiversity, the importance given to beekeeping and the industrialization of its products, as an economic and sustainable activity, is not remarkable until now. The geographical location of Colombia, in the equatorial zone of South America, implies the inexistence of seasons and a permanent flowering linked to a great variety of plant species, giving high potentials of honey production compared to reported by other countries. Likewise, the diversity of ecosystems entails a variety of products according to the geographical origin, an aspect that has not been considered until now. The composition of honey allows evaluating its quality, which could change according to the botanical origin, geography, climate, time of harvest, techniques used during the extraction, among others. However, the lack of legislation has caused a high index of falsification and poor verification of the quality and differentiation. Based on the above, the objective of this study was to characterize and classify 115 samples of honey from

23 different political zones of Colombia, through the identification of a particular fingerprint based on physicochemical and instrumental sensory properties. The evaluated parameters were pH, free acidity, diastase, ash, hydroxymethylfurfural, color, electrical conductivity, and moisture. Moreover, it was employed an electronic nose with 10 non-specific metal oxide semiconductor sensors for evaluation of the aromatic profile (volatile components). Finally, three electrochemical sensors screen-printed were used to measure the presence of electroactive species in solution (non-volatile components), by a voltammetry technique. The data were analyzed by unsupervised and supervised methods such as principal component analysis (PCA) and Discriminant Analysis (DA). This study found evidence that the samples complied with the main parameters of quality and composition reported in the local and external standards. Discrimination by origin of honey allowed grouping the samples according to the geographical origin. This study aims to contribute to the recognition and differentiation of physical-chemical characteristics of honey according to their origin, as well as to recognize the products originating in this country abroad, in order to increase their commercial value.

[P.11.275] Antioxidant activity of proteic fraction of honey of *Melipona beecheii* and *Apis mellifera* bee

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The excess of free radicals in humans is related to some diseases such as cancer, diabetes, cardiovascular problems, Alzheimer, among others. Several environmental factors such as UV, pollution, smoke, stress, etc., trigger the production of free radicals. Recently, it has been found that extracts of natural proteins or purified proteins have antioxidant activity, such as isolated soy protein, corn zein, potato and chickpeas proteins. These owe their activity to the amino acids that constitute them. Honey is a functional food, has biological properties such as antibacterial, anti-inflammatory and antioxidant, which depend on its composition. Its antioxidant activity is due in part to the polyphenol content and probably to the protein and peptide content. In the Yucatan peninsula, Mexico, bee honey is produecd from two species of bees, an autochthonous one *Melipona beecheii* which are characterized by having no stings and another *Apis mellifera* which were introduced during colonial times. The objective of this study was to determine the antioxidant activity of the protein fraction of bee honey from multifloral *Melipona beecheii* (10 samples) and *Apis mellifera* from two flowering Tajonal and Tzitzilche (18 samples), with two methods, DPPH and ABTS. The total average protein content of the samples was 0.78% for Melipona honey and 0.70% for Apis honey. The antioxidant activity evaluated with the DPPH method was 0.096 mg EQ / 100 g of honey for Melipona honey and 0.079 and 0.135 mg EQ / 100g of honey, for honey of bee Apis from Tajonal and Tzitzilche flowering respectively. With ABTS method, the antioxidant activities were 0.107, 0.058 and 0.067 mg EQ / 100g of honey, respectively. With DPPH method antioxidant activity of protein fraction of melipone honey was higher than Apis and with ABTS method that of Tzitzilche honey was the highest. These results suggest that the composition of the proteins of the three types of honey must be different.

[P.11.276] Brazilian organic honey has high capacity to deactivate reactive oxygen and nitrogen species

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Honey is an easily accessible honeybee product, increasingly popular due to its biological activities and beneficial effects on human health. This study aimed to evaluate the capacity of extracts of five organic blossom honey and three honeydew honey from southern Brazil to deactivate reactive oxygen (ROS), as well content of vitamin C. Total polyphenol contents (TPC) were determined using the Folin-Ciocalteu method (Singleton, Orthofer, & Lamuela-Raventós, 1999) adapted to a microplate reader an all the honey extracts analyzed displayed high content of phenolic compounds (49.79–117.68 mg gallic acid /per gram of extract). Peroxyl radical (ROO•) scavenging capacity was determined according to Melo et al. (2015), and the honey extracts analyzed displayed high capacity to deactivate peroxyl radical (ROO•) (835.54 - 1785.35 µmol Trolox/ per gram of extract). Determination of ascorbic was using HPLC-DAD coupled online with ABTS analysis followed an adaptation of a method previously used by our team (Tiveron et al., 2016). Honeydew honeys exhibited high contents of ascorbic acid (2.75 to 6.22 mg/100 g honeydew honey) accounting for their antioxidant activity. Therefore, the organic honeys produced in southern Brazil can be considered good sources of bioactive phenolic compounds, and if consumed on a regular basis, they can contribute for decreasing excessive ROS, responsible for harmful effects to the structural and functional integrity of cells.

[P.11.277] Protective effect of south Brazilian organic propolis against reactive oxygen species

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The aim of this work was to evaluate the protective effects of Brazilian certified organic propolis against reactive oxygen species (ROS) such as peroxyl radical (ORAC), superoxide and hypochlorous acid. This biological activity has been attributed mainly due to the presence of phenolic compounds and/or derivatives. However, chemical composition of propolis depends on the vegetation around the hive. The

propolis collected in permanent preservation and reforestation areas (78 samples) were classified in seven chromatographic profiles (OP 1-7), according to the HPTLC analysis. The OP1 showed the highest ORAC value (1.95 µmol Trolox/mg) and PO6 the lowest one (0.5 µmol Trolox/mg). IC50 values required to inhibit 50% of superoxide anion radical ranged from 0.29 µg/mL to 2.91 µg/mL. Profiles of organic propolis also showed significant results regarding the hypochlorous acid scavenging. All profiles, except OP 6, showed similar IC50 values. These results demonstrated that organic propolis has important antioxidant activity, since many diseases are associated with ROS.

[P.11.278] Effect of Skin care cosmetic of honeybee (*Apis mellifera*) dron extract by test of Physiological Activities on cell

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This test was conducted to evaluate the effect of the in vitro wrinkles, whitening and moisturizing of the honeybee pupa extract of western species. 100ug/ml, which is not toxic to both HDF and B16F10 melanoma cells, was set at its highest concentration and 20 and 4 ug/ml were set at its test concentration with a ball ratio of 1/5. The amount of collagen type I in UVB-tested cells treated with extract increased with higher concentrations of the sample, and the expression of the MMP-1 decreased with higher concentrations. The results of the melanin-producing inhibition test of the extract indicated that melanin tends to decrease when the extract is processed. In addition, the results of the in vitro tyrosinase incubation test using L-Tyrosine and L-DOPA as substrates showed that the effect of tyrosinase inhibition was increased to 40.7, 62.5, 83.8%, and 53.4, 57.7, and 61.9%, respectively, when the substrate furnace was used in the , to control the tyrosinase when the concentration was high. The results of the investigation of the expression of Fillaggrain and total cholesterol content in TGM1 and HDF cell showed no difference between control and treatments sample. In conclusion, It is judged that the water bee pupa extract increases the collagen production, suppresses the expression of the collagen degradation enzyme MMP-1 and has effect on whitening. However, it does not have a moisturizing effect.

[P.11.279] The antioxidant and antimicrobial properties of Meliponinae honey Collected on Different Days

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Stingless bees have degraded wing, well-developed mandibles, no stings, and pollen baskets on the hind foot. The size of stingless bee are one-tenth to western bees, mainly found in tropical and subtropical regions such as Australia, Africa, Southeast Asia., North America and South America. However different species can produce honey with different content of phenols and flavonoids, and have different effects on antioxidant, antimicrobial and anti-inflammatory activities. This experiment explored *Heterotrigona itama* and Tetragona binghami honey in Malaysia have different effect on physicochemical properties, antioxidant and antimicrobial. In the result showed that the acid increase with harvested day the organic acid content also has tendency to increase. In the phenols and flavonoids content result show that both the Itama and binghami products had tendency to increase. In the antioxidant activity (DPPH free radical scavenging, FRAP and reducing power) results showed Itama antioxidant capacity better than binghami and had the best antioxidant activity for 45 days. The antimicrobial activity test results showed that due to the higher acid value and low pH value of Tetragona binghami, it also showed a better antimicrobial profile in antimicrobial results. According the results, it can be seen that the physicochemical properties, antioxidant activity and antimicrobial activity are different depending on the type of Stingless bee species and honey collected on different days.

[P.11.280] The Addition of Flower Pollen to Natural Gum

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Natural chewing gum is a thick and sticky gum which is antibacterial and effective in healing wounds and injuries. Flower pollen is a product of bees and with essential amino acids, vitamins, minerals and hormones is useful for people with a weak nervous system. Research has shown that flower pollen with vitamin B is a complex immunosuppressant and it has natural antioxidants. It is also effective in the treatment of diabetes. Accordingly, the aim of this research is to add pollen to natural gum and the study of its physicochemical properties.

In this study, it has been tried to avoid the presence of artificial sweeteners with the inclusion of flower pollen in the natural gum formula. In a study by researchers in 1997, a new production of pollen from flowers and propolis has been explored which can control the amount of sugar and fat. This product softens the blood vessels, improves blood circulation and prevents complications of diabetes and even improves it.

In another study conducted in 1999, the combination of flower pollen with propolis, royal jelly and honey was effective in treating prostate disease. It should be noted that chewing gum plays an important role in digestion by activating the cerebral cortex and improving thinking and consciousness and better processing of information and also secretion of saliva. But synthetic types of it are non-degradable latex and plastics in the digestive tract and in the environment. The release of this small hazardous waste causes a variety of environmental and water pollution.

Accordingly, the results of this study show that producing natural gum with pollen, in addition to being an environmentally friendly product,

it is easily digested in the gastrointestinal tract and with flower pollen instead of sucrose and all kinds of artificial sweeteners, it can be useful in the treatment of diabetes and diseases like prostate. Results of organoleptic test and general acceptance in relation to taste as well as natural gum tissue show that this chewing gum has been accepted by the consumer.

[P.11.281] Spray-dry optimization of *Melipona beecheii* honey powder

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In order to preserve funtional and nutricional properities and to extend shelf life of stingless bee (*Melipona beecheii*) honey, a spray-dry optimization study was performed to obtain a product of honey powder. Honey samples were collected from melipona beehives from Calkiní, Campeche, México. A statistical methodology of response surface was carried out in order to achieve the best conditions of spray-dry process to obtain a stingless bee honey powder. Independet variables were inlet air temperature (50-70 %) and maltodextrin (DE 10) content (50-70 %). Response variables were as follows: powder yield, humidity, volatile compounds retention, solubility and hidroximethylfurfural content (HMF). Response surface statistic analysis yield the optimal operating spray-dry conditions at inlet air temperature 150 oC and maltodextrin 68%. Reological properties of honey powders at optimal conditions were as follws: yield 45%, humidity 4.9%, volatile compounds retention 77%, solubility time 255 s and HMF content 52 mg/kg.

[P.11.282] Physicochemical quality of the honey of *Apis mellifera* Africanized produced in soybean growing area – Brazil Southern

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Honey is a natural product consumed in all world, and new consumer markets are being reached considering its nutraceutical properties. The characteristics vary mainly according to the floral source, due to its complex composition. This study aimed to evaluate the physicochemical profile of honey samples obtained from two soybean cultivation areas: open field (provided by a beekeeper) and closed plot in a 24 m² (4m x 6m) pollination cages. The evaluated parameters were water insoluble content, protein, acidity index, pH, moisture, fixed mineral residue and composition of sugars. Qualitative and quantitative identification of pollen grains present in the honey (palynological analysis) is a tool to define the identity and authenticity in relation to its floral origin. The honey sample from open field was submitted to the palynological analysis. The chemical composition of the grains of soybean pollen was done by X-Ray Diffraction (XRD). Therefore, the honey sample obtained in the closed plot (soybean flowers as unique protein and energy source) presented higher acidity and lower protein contents, when compared with that from open field. The palynological analysis showed that Eucalyptus (Myrtaceae) was the most representative source (38%) followed by Leucaena leucocephala and Glycine max, (both Fabaceae) and Baccharis sp. (Asteraceae) with 20, 19 and 16% respectively. XRD analysis demonstrated the following composition: C, O, Ca, K, P, Mg and S of the soybean pollen present into honey sample. The physicochemical profile from both, open field and closed plot, honey samples are in agreement with the current rules of the Mercosur (GMC 89/99) and Brazil (Normative Instruction, nº11/2000), regarding characterization of floral honey. The palynological analysis of the open field sample demonstrates a greater visitation of the honeybees in eucalyptus. As 19% of the pollen grains are of soybean origin, it is a clear confirmation of the foraging of the honeybees in this crop of great economic importance, evidencing the interest of honeybees in soybean plants. This is a finding that goes against the common sense that soybean plant, for it is autogamic and cleistogamic behavior, does not attract pollinator's agents.

[P.11.283] Physicochemical composition of *Apis mellifera* drone pupae

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Drone pupae of honeybees (*Apis mellifera*) have been used as food for enhancing immune system in many countries. However, despite its high nutritional value, it is not registered as food ingredient in Korean food standards codex. In the present study, we evaluated the nutritional characterization of drone pupa through chemical analysis. The contents of moisture, crude protein, crude fat, carbonate and ash in proximate composition were 74.23%, 11.05%, 8.19%, 5.68% and 0.85%, respectively. 18 amino acids, including 9 essential amino acids were found in drone pupa with the highest level of glutamic acid at 1631.9 mg/100g. The drone pupa which contains 12 minerals was rich in K (235.78 mg/100 g) and P (177.35 mg/100 g). Vitamins (B1, C and E) and fatty acids were present at low concentrations. These results suggest that honeybee drone pupa is a rich source of protein and other essential nutrients and thus could be used as a food ingredient.

[P.11.284] Evaluating the levels of sugar, phenol compounds and Vitamin C in Single sunflower honey and several flowers honey

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Natural honey tends to be one of the important materials in international markets, while it is a profitable matter for many countries, financially. Chemical composition of honey and its specific nature is different based on climate and vegetation of the very areas. The aim of this study is to evaluate the physiocochemical and biochemical nature of the Single sunflower honey and several flowers honey. Therefore, 10 samples of several flowers honey from areas of Sero and 10 samples of sunflower honey from rural areas of Anganeh village in Urmia were collected and studied. Results depicted that the average of Revitalizing sugars along with phenolic substances were higher in several flowers honey ($P<0.05$). However, there were no significant difference between the total sugar of Single sunflower honey and several flowers honey ($P>0.05$). The average composition of Single sunflower honey and several flowers honey included 77.78±3.5% and 77.1±1.74% sugar, 5.31±1.11% and 3.53±0.83% sucrose, 83.37±3.8% and 85.84±2.4% total sugar, respectively. Besides, Single sunflower honey and several flowers honey included 26.4±9.3 and 25.5±2.68 mg/100g of vitamin C, and 0.017±0.01 and 0.033±0.008 (absorption in 765 nm) of phenolic substances. The results of this study indicated that the amount of sucrose and Revitalizing sugars of both of the honey are not compatible with the international ones. However, other substances of both honey were comparable with other honeys, worldwide, and they were in the range of international standards.

[P.11.285] Propolis extract for preventing weed seed germination as a natural herbicide

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University of Tehran, Karaj, IRAN

Propolis is one of the most valuable bee products that have many applications including medicine, agriculture and animal husbandry. In this study, a new application has been made regarding the effect of propolis in agriculture. Weeds annihilate a large part of agricultural crops. Therefore, many expenses for controlling and destroying weeds are spent on the purchase of herbicide. They cause environmental pollution and have side-effects to human. For the production of herbicide, propolis extract was used in four concentrations. Also, the combinations of propolis extracts were tested on germination of wild barley, oat and cattle cotton. For primary treatment, the seeds were grown in Peat Moss and each day 2 ml of the specified concentrations of the extract of the propolis were injected. Three replicates for each treatment and three replicates without treatment were considered as controls. All treatments and control were irrigated daily. After determining the best concentration in preventing germination of seeds, the duration of starting germination after application of this extract in the ground was investigated. For this purpose, germination of wheat was studied after application of the extract 1: 2 in culture medium at 1, 8, 16 and 24 hours after treatment. The results of primary treatment showed a significant effect of propolis extract compared to the control. Also, the results of the secondary treatment showed that this extract does not prevent germination of seed of the main plant after soil treatment. In conclusion, this extract can be recommended as a herbicide before cultivation on farms.

[P.11.286] Bee Products for Technology and Marketing

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Forestry Research Institute of Ghana, Kumasi, GHANA

The bee farming initiatives aims to encourage small, medium and large scale farmers in the tropics to consciously integrate bee farming into their cropping systems to increase crop yields and biodiversity conservation through pollination services and to provide incomes from the sale of beehive products(honey, beeswax, propolis). As the population is increasing in the tropics, land availability and ownership by farmers mostly who are women are decreasing just as yields are also decreasing due to over over dependence on inorganic fertilizers and pesticides with no due attention to bees which constitute 60% of pollination services for increased crop yield and alternative revenue to farmers. In summary the benefits of integrating bee-farming into the crop production system are as follows. Honey, beeswax, propolis and other beehive products which are cash crops' are produced with less investment in terms of fertilizes irrigation pesticides etc. Beekeeping has positive ecological benefits because about three-quarters of crops worldwide are pollinated by bees(both sting-less and stinging bees). Less land size is used to obtain high revenues and the honeybee does not compete for resources with any other agricultural enterprises. Forestry Research Institute of Ghana is putting measures in place in order to create the awareness that bee farming (Beekeeping) is a livelihood option which can be depended on by breaking the myth surrounding the fear of bees through education, film shows and demonstration.
Development of low-technology beehives using lesser used wood (e.g coconut lumber, basket and palm branches mid-ribs) to reduce initial investment cost to maximize income. Also, promoting both orthodox and traditional medical benefits of honey, beeswax, propolis and other beehive products using printed brochure. Identification and incorporation of various plant species into the farm for all year round nectar and pollen flow. Mastering queen breeding and bee multiplication to have more colonized hives without depending on bee colonization by nature and improving on aggressive marketing methods.

[P.11.287] Encenillo honey as unique monofloral honey reported in Colombia

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Colombian honey possesses large floral sources with various colors and flavors, mainly due to the differential floral offer at the wild life areas, which depend on the altitudinal conditions and their botanical and geographical differences. Encenillo honey is a natural product with sweetening properties produced by bees from the nectar of flowers and secretions of Weinmannia tomentosa Don., at highlands Andean zones in Boyacá, Colombia. The aim of this work was focused on physicochemical properties produced by Encenillo from highlands Andean in Colombia, that were characterized by melissopalynology, sensory attributes, volatile compounds as chemical composition. Pollen grains at the samples were greater than 45%, conferring a quality of monofloral. Color pfund 69.0 ± 1.00, pH and total acidity 3.87 ± 0.04; 31.8 ± 1.40 meq kg-1, still within the standard limits, electrical conductivity 0.525 ± 0.004 mS cm-1, moisture 20.1 ± 0.43 g 100 g-1, ash 0.198 ± 0.002%, reducing sugars 69.7 ± 0.17 g 100g-1, total phenols 1570 ± 14 mg kg-1 G.A. flavonoids as Quercetin 129.4 ± 1.20 mg kg-1. Tabouret's index 3.75 ± 0.06 based on 1.35 ± 0.03 relationship fructose/glucose and 0.604 ± 0.050 water activity. Honey show a Newtonian's behavior. GC/MS analysis permitted to identify 25 volatile compounds, between alcohols, aldehydes, ketones, esters, hydrocarbons, furans, nitriles, sulfide and terpenes. The study is related as a model for characterization of types of honey and consolidated as the first research on unifloral honey in Colombia.

[P.11.288] The influence of protection procedure of geographic indication of honey in Serbia on quality improvement of bee products

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In the period from 2008 until today, on the territory of the Republic of Serbia, 5 types of honey with a geographical indication have been protected. Two honey, Homolje honey and Fruska Gora lime tree honey have a protected name of origin. Others, Kacer honey, Djerdap honey and Vlasina honey have protected geographical indications. Thanks to the increased interest of beekeepers in Serbia to increase the additional value of honey, it protection has started. With the help of the Ministry of Agriculture of the Republic of Serbia, the Institute for Intellectual Property of the Republic of Serbia, and the experts for performing tasks related to the protection of the geographical origin of honey, the work has started and is still ongoing. Protection of Deliblat honey is in progress, and protection of honey from Stara Planina and several geographically defined areas is planned, where a quality honey is obtained with characteristic organoleptic properties and special pollen formulas.
All honey protection activities had a significant impact on the increase in the purchase prices of honey originating from areas that were protected. Also, there is noticeable effect on beekeepers's awareness to produce high-quality, health-safe products and to reduce the quantity of falsified honey in the territory of the Republic of Serbia.

[P.11.289] Effect of processing temperature on biochemical components in honey of European honeybee, *Apis mellifera* from the northern Thailand

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King Mongkut s University of Technology Thonburi, Ratchaburi, THAILAND

Honey is the best bio-sweetener in nature consisting of enzymatic activity, antioxidants, and nutrients. However, the temperature can alters those essential components along honey processing and storage life cycle. This study aims to analyze biochemical composition of honey samples obtained from the north of Thailand. An activity of enzymes both invertase and diastase and quantity of hydroxymethylfurfural (HMF) are considered as the parameters. Honey sample has been heating under three heating temperatures (45oC, 55oC and 65oC) for 1, 3 and 5 hours duration. The results showed that an increase temperature cause a decrease enzyme activity. At 45oC of incubation time 1, 3 and 5 hours, invertase activity of sample was decreased 3, 12 and 19 times while diastase activity was decreased 9, 20 and 18 times compare to original honey sample. Hydroxymethylfurfural (HMF) was increase 36, 62 and 67 times at heating duration of 1, 3 and 5 hours, respectively. However, at heating temperature of 55 and 65oC for 5 hours, enzyme activity had the drastically reduced to 90% and HMF had increased quantity more than 200 times. The comparison of biochemical parameters between capped and uncapped honey processes at 45oC for 1 hour duration showed no significant different in of which found reduction of activity about 10-15% in invertase and 12-18% in diastase. On the other hand, HMF was rapidly increased more than 50%. The results of this study indicate that the temperature significantly affects the biochemical component on honey of *Apis mellifera* from the north of Thailand.

[P.11.290] Evaluating the amount of humidity, sucrose, insoluble solid compounds, Brix, density, phenol compounds and vitamin C in Single sunflower honey and several flowers honey

A. Hazrati [1], M. Shahi Bilesavar [1], M. Ghasemi Heidarlu [2], M. Soraiaband [3]
[1] *Islamic Azad University - Ph.D. student of Food Hygiene, Tabriz, IRAN,* [2] *Board of Beekeepers Union, Urmia, IRAN,* [3] *Site design company, Urmia, IRAN*

Natural honey tends to be one of the important materials in international markets, while it is a profitable matter for many countries, financially. Chemical composition of honey and its specific nature is different based on climate and vegetation of the very areas. The aim of this study is to evaluate the physicochemical and biochemical nature of the Single sunflower honey and several flowers honey. Therefore, 10 samples of several flowers honey from areas of Sero and 10 samples of sunflower honey from rural areas of Anganeh village in Urmia were collected and studied. Findings depicted that the average of humidity and sucrose were higher in Single sunflower honey, while the average of density, phenol substances, and insoluble solid substances were higher in several flowers honey ($P<0.05$). On the other hand, there were no significant differences amongst the brix and vitamin C of the honeys ($P>0.05$). The average of the Single sunflower honey and several flowers honeys included 15.51±0.44 and 14.22±0.42% humidity, 1.062±0.001 and 1.066±0.001 density, 26.9±4.3 and 25.5±2.68 mg/100g of vitamin C, 0.017±0.01 and 0.033±0.008 (absorption in 765nm) phenol substances, 0.56±0.18 insoluble solid substances, and 77.1±1.74 and 78.0±1.74 brix. These results indicated that the sucrose amount of both of the honeys are not compatible with the international ones. However, other compositions in both honeys were comparable with other honeys, worldwide, and they were in the range of international standards.

[P.11.291] A Review on Crude Beeswax Mismanagement and Lose: Opportunities for Collection, Processing and Marketing in Ethiopia

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Beeswax is a valuable product that can provide a worthwhile income in addition to honey. One kilogram of beeswax is worth more than two folds of a kilogram of honey. In Ethiopia, beeswax is one of the important exportable agricultural commodities; the annual production of beeswax is estimated to be more than 5,000 tons. About 64,000tons of beeswax were produced in the world, Asia (mainly India) being the major producer with 31,000 tons. Of all honey bee products the economic importance of beeswax is second after that of honey. Tropical countries dominate world beeswax production and export, with industrialized countries needing to import beeswax. Ethiopia is among the top four beeswax producers which is attributed to the predominantly traditional system of beekeeping production. The valuable beeswax resource is neglected in some areas of the tropics. Crude beeswax is collected from three sources. Of the total production the major part is utilized for the production of candle, and Twaf, a candle like stick that used for church ceremony. Establishment of a beeswax collection and processing centre with the accessible rendering technologies and encouraging the beekeeper to handle the beeswax resource. Beeswax is relatively expensive, and there has always been a tendency for people to falsify or dilute with cheaper materials.

[P.11.292] Simple uplc-esi-ms/ms method for rapid simultaneous multi-class determination of antibiotics residues in honey

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Honey is widely consumed as food and dietary supplement. According to EU regulations, honey is considered as a natural product and must be free of contaminants. Beekeepers use antibacterials to prevent and treat bees' infections, but the use of most antibiotics is illegal in many EU and American countries. Such chemicals can persist at trace levels in honey for a long time and may have toxic effects on consumers' health. Despite for the ban, RASFF database during 2002-2018 has showed large number of notifications for antibiotics in honey, indicating that honey contamination is still present. So antibiotic residues determination is an obligatory demand for honey import and export globally.

Multi-class and multi-residue analytical protocol of antimicrobials determination in honey is necessary to accelerate the process of control of drug residues in more cost-effective way. So we have developed and validated simple, sensitive and rapid UPLC-MS/MS method to detect and quantify six classes of most common antibiotics in honey, viz. amphenicols, nitroimidazoles and their metabolites, sulfonamides, trimetoprim, tetracyclines, fluoroquinolones and nitrofurans (NF) metabolites (~30 analytes). Main steps of sample preparation are as follows: acid hydrolysis and ultrasonication of bonded analytes with simultaneous derivatization of NF metabolites, neutralization by buffer solution with following extraction by ethyl acetate and reconstitution of evaporated extract in mobile phase. Target compounds identification, quantification and confirmation are carried out in MRM mode. Proposed analytical technique is capable for multi-class selective determination with LODs from 0.05 ppb for chloramphenicol to 2 ppb for tetracycline using matrix matched calibration. The evaluation of procedure's adequacy has been carried out according to 2002/657/EC requirements and by inter-laboratory testing.

Developed method has sufficient advantages comparing with known multi-residue LC-MS/MS methods, viz. it does not demand the use of expensive and time consuming solid phase extraction procedure without a loss of sensitivity and selectivity. But the main benefit of our

method is an ability to determine NF simultaneously with sulfonamides and tetracyclines. Hence, developed multi-analytical UPLC-MS/MS technique is fast, robust, reliable and cost-effective. This method is routinely used in our laboratory to test export lots of Ukrainian honey and for incoming control.

[P.11.293] Lithium chloride - hazard or possibility?

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Ectoparasite Varroa destructor is recognized as the biggest pest to the western honey bee worldwide. Left untreated, mites can kill an entire colony within an apicultural season. Controls are mainly effective, but restricted only to few chemicals implicating the risk of evolving resistance posing mite eradication unpredictable in the foreseeable future.

Recently, alongside of novel approaches as RNAi to control varroosis it was found that Lithium chloride may be a promising and easy-to-use chemical to effectively treat Varroa infestation. Within days of publication of its potential to eradicate the Varroa mite, beekeepers started to buy commercially available lithium chloride and started to apply it in private apiaries without being aware that the use of LiCl may have unwanted impacts on human exposure though the food chain.

Having psychotropic properties, lithium salts are extensively used in manic-depressive disorders treatments. However, therapeutic doses are close to the maximum of the intake level. Whether the use of LiCl in apiculture as treatment against Varroa infestation will pose a risk of hive products becoming polluted or not is still unknown. It may affect any apicultural product since therapeutically fed lithiated sugar syrup is eaten by the bees.

The aim of the present study was to reveal the accumulation of lithium-chloride in different apicultural products, involving also adult honey bees, their brood as well.

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BEEKEEPING TECHNOLOGY AND QUALITY

11 SEPTEMBER 2019

POSTER SESSION 16 08:30-18:00

BEEKEEPING TECHNOLOGY AND QUALITY II POSTER AREA

[P.16.371] Discrimination of geographical origins of Chinese acacia honey using complex 13C/12C, oligosaccharides and polyphenols

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The aim of this study was to predict the geographical origin of acacia honey of China through analysis of physicochemical parameters combination with chemometrics. Samples from six different origins were investigated on parameters of stable carbon isotope ratio value, oligosaccharides and polyphenols, using EA-IRMS/LC-IRMS, GC–MS and HPLC–MS, respectively. The results indicated that the stable carbon isotope ratio value of honey from Gansu region were lower than those of other regions. Oligosaccharides of honey from Shanxi and Shaanxi regions were both higher than other four regions. Polyphenols of honey from Shandong region was the highest and were better parameters than both stable carbon isotope ratio and oligosaccharides in discrimination of geographical origins. Partial Least Square Discriminant Analysis (PLS-DA) showed that when all 31 different parameters were combined, a correct classification rate of 94.12% could be achieved using external cross validation method. In conclusion, the method in discrimination of geographical can be used to provide reliable and useful reference information.

[P.16.372] NMR Honey-Profiling™ Reloaded – Improvements in the 2nd Version

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In 2015 the first release of NMR Honey-Profiling™, which QSI co-developed with Bruker BioSpin, Rheinstetten, Germany, and partners, was put into action. Though already powerful, the database at that time contained little less than 4000 samples and only a small number of verification models for few geographical and even fewer botanical origins. For the detection of adulteration 15 models were available.
Since October 2018 the second release is activated offering a number of highly valuable improvements. This new version of NMR Honey-Profiling™ contains more than 18.000 samples of worldwide origins, which makes it the biggest database for honey and syrups worldwide. Each sample that was added to the database went through a thorough check-up using existing (conventional) methods, e.g. AOAC 998.12 (13C-isotopes), analysis of the pollen spectra, foreign enzymes, oligosaccharides and others, in order to assure authenticity of the samples.
The new release now offers 60 models for adulteration, 20 models for verification of the geographical origin and 12 models for the verification of botanical origins.
From the technical point of view, results of Honey-Profiling™ were always highly comparable between different laboratories. However, when using the first version, in some cases different results were submitted to clients due to the subsequent (and absolutely necessary) expert interpretation of the results, which may differ from lab to lab. The newly introduced dedicated models helped harmonizing evaluation between laboratories, as a possible misinterpretation of deviations from authentic honey is far less likely with the new version of NMR Honey-Profiling™. Examples will be provided in the presentation.
Through the expansion from 15 to 60 adulteration models it was possible to increase the detection rate to nearly 75% of adulterations, which is far more than any other method can detect to date. The remaining ~25% of undetected adulterations can be mostly attributed to parameters detected using highly sensitive methods like foreign enzyme activities, which cannot be detected using NMR so far. However, development proceeds continuously to further increase the rate of detection of adulteration using Honey-Profiling™.

[P.16.373] Evaluation of Commercial Korean Honey Quality and Correlation Analysis of the Quality Parameters

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Honey is made from flower nectar by honey bees. In this study, 120 honeys from various flowers and across eight different provinces in Korea were collected and their components, antioxidants, and hemolytic activities against red blood cell were evaluated. Our results show that total polyphenol (TP) varied widely across the samples, with chestnut honey showing the highest TP (77.1±8.4 mg/100g), protein content (25.9±0.9 mg/100g), and absorbance at 400 nm (A400 : 0.156±0.036). In contrast, the acacia honey and sugar honey had a TP of 9.5~30 mg, 12~15 mg/100g of, and the lowest A400 of 0.06±0.02. High amounts of total flavonoid were quantified in the jujube and chestnut honeys at 8.73±7.31 and 8.39±3.02 mg/100g, respectively. No samples demonstrated hemolytic activity up to 1 mg/ml.

Antioxidant activities determined by DPPH, ABTS, and nitrite scavenging placed the chestnut honey highest, followed by jujube, styrax, multi-floral, citrus, acacia and sugar honey. Analysis of parameter correlations indicated that the components and bioactivity of the honey are dependent on the origin of the flower rather than on bee-farming regions. A positive correlation between TP content and antioxidant activity was identified. The correlation coefficients between A400 and the TP, ABTS scavenging, and reducing power values were 0.804, 0.772 and 0.741, respectively. We therefore suggest that A400 could be used as a noble, economic and simple factor for honey quality evaluation. Our results can potentially be used to develop functional honey for the food and pharmaceutical industries.

[P.16.374] How trace elements reveal adulteration of honey

C.C. Luellmann [1], A. Duebecke [2, 3], T. Wiezorek [4], G.I. Beckh [2]
[1] *Tentamus Group Gmbh, Berlin, GERMANY,* [2] *Quality Services International Gmbh, Bremen, GERMANY,* [3] *Tentamus Center For Food Fraud - Tcf2, Bremen, GERMANY,* [4] *Qsi America / Adamson Analytical Laboratories, Corona, USA*

About three decades ago QSI started to look into trace elemental composition of different honeys worldwide. However, the complexity of this multi-parameter method and the necessary computing power needed for statistical evaluation rendered this method as not practicable at that time and focus was set mostly on methods targeting fewer parameters.
Nowadays computing power is not an issue any more and interest in this approach is increasing again. In a recent publication in Nature Magazine the options that this approach is offering are highlighted again. US authorities also have recognized the chances that this method provides to check imports especially from Asian countries for adulteration.
In order to be able to use the trace elemental composition to detect adulteration, it is necessary to build up a database of samples from the countries that you want to compare. Through the international network of QSI it was possible to collect samples from all over the world. These samples are used for the development of a method based on trace elemental composition of honey and syrup in order to provide a means for importers to test their honey before import.
We analyzed the trace elemental composition of honey samples and syrup samples from different countries with significant export volumes and focused on adulteration. A broad range of different major and minor trace elements was targeted in order to give a comprehensive overview on these samples and to maximize the detection rate of adulteration.
In this presentation the results of our study will be provided and chances but also limitations of this approach will be discussed.

[P.16.375] The MALDI-BeeTyping, an innovative method to monitor the quality of bee products

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Bee products are increasingly used for their functional properties in human and animal health. Some of these products reach high prices in the market and are subject to more and more frauds (e.g. kanuka honey vs manuka honey, bulk imported royal jelly vs local jelly with good practices).
We established the profile of different bee products based on MALDI-BeeTyping, an approach derived from the BioTyping used in clinical microbiology laboratories, and already adapted to bee health diagnosis in our laboratory. This proteomic barcoding technique gives a molecular profile of the bee product by MALDI TOFF Mass spectrometry.
We generated statistical models for different bee products (e.g. Manuka honey, royal jelly with quality sign, propolis) and compared them to models of imported or improper products found on the market. We discuss the possible use of this reliable, cost effective technique as a new tool in the quality control of honeybee products.

[P.16.376] Survival of a special probiotic bacteria in honey: during storage and simulated gastrointestinal conditions

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[1] *Armaghanbehshahd Chichest (mirnajmi Honey Co.), Urmia, IRAN,* [2] *Research Institute of Food Science And Technology, Mashhad, IRAN*

There is strong evidence documenting the specific promotion effects of probiotic microorganisms on intestinal tract and overall health. Within the last decade, development of non-dairy based probiotic products which need no specific equipment for cold keeping, have experienced rapid marketing growth. Honey as a natural and popular sweetener contains a lot of nutritional ingredients and could be an appropriate carrier for delivery of probiotics to the human body. The success in production of non-dairy probiotic products could be influenced by selecting the suitable probiotic strains. In the present research, honey was enriched with a special probiotic bacteria which has a great resistance to stomach acid and harsh processing conditions used in the food industry (107 cfu/g). The viability of probiotic bacteria during processing and storage was evaluated. Moreover, the survival of bacteria cells in probiotic honey was investigated after passing the simulated gastrointestinal conditions (SGC). According to the results, the probiotic bacteria reduced from 107 to 2×106 cfu/g after 6 months of storage time. The obtained results showed the protective effect of honey on probiotic bacteria while passing the SGC. It has to be mentioned that the physicochemical properties of probiotic honey including pH, acidity, color, reducing sugars, sucrose, fructose/glucose ratio, proline, hydroxymethylfurfural, osmophilic yeast and mold counts were at the standard ranges. These results suggested the honey as a novel, appropriate medium for fortification with probiotics which could positively affect its global marketing.

[P.16.377] Yeast as an indicator of honey's authenticity

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[1] *Slovenian Beekeepers Association, Lukovica, SLOVENIA,* [2] *Jozef Stefan Institute, Ljubljana, SLOVENIA,* [3] *University of Ljubljana, Biotechnical Faculty, Ljubljana, SLOVENIA*

Honey has always been a highly valued food product because it has great nutritional value and helps maintain and improve people's health. Due to the expensive production, it is often a target of adulteration, both intentional and unintentional, as a result of the beekeeping technology. The beekeepers must feed the bees before winter or during their non-foraging period, this feed can later be found in honey, also because bees carry it around the hive. There are several methods of checking the honey authenticity. If a beekeeper adds yeast to bees' feed, yeast may appear in the honey. In the springtime we fed bee colonies (18 colonies) with honey-bee candy, which had added yeast in them. After the honey extraction, we compared the results of the number of yeasts found in the honey with the results of physicochemical parameters (electrical conductivity, water content, free acids, diastase number), foreign enzymes in honey and stable isotopes. Yeast can be a good indicator of honey adulteration; they were strongly correlated with the presence of foreign enzymes in honey.

[P.16.378] In-vitro antimicrobial properties of bee products

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Bee products have been extensively used in treatment of various diseases in folk medicine since ancient times. A number of investigations have shown that bee products possess antibacterial, antiviral and antifungal properties. In recent years, interest in natural medicines and bee products has increased due to the side effects of medicines.
In this research in vitro antibacterial activity of 8 different honey and 10 different propolis products were tested against 5 Gram-positive bacteria, 7 Gram-negative bacteria, Mycobacterium smegmatis and yeast.
Drops (15%, 30% propolis), spray (6% propolis) and raw honey mixtures (1%, 2%, 10% propolis) were tested by agar well diffusion and inhibition zones were compared.
As a result of the study, the products in the form of drops inhibited all tested Gram-negative and Gram-positive bacteria species. A significant antimicrobial activity has been observed especially against Helicobacter pylori, Staphylococcus aureus (20-18 mm), Bacillus cereus (15-18 mm) and Mycobacterium smegmatis (19-18 mm).
The product in spray form showed antimicrobial activity against 12 of the tested bacteria and yeast species. Measured inhibition zone diameters varied between 6-45 mm.
The propolis-raw pine honey and propolis-raw chestnut honey mixtures which contains 10% propolis demonstrated antimicrobial activity against 12 different bacteria and yeast species. In propolis-raw honey mixtures, the zone diameters were in the range of 6-30 mm.
Turkish astragalus, pine, chestnut and Hakkâri region honey and 4 different manuka honey also were compered against ten different microorganisms. It has been observed that pine honey is as effective as manuka honey against Staphylococcus aureus. The antimicrobial activity of the chestnut honey against Escherichia coli, Citrobacter freundii and Bacillus cereus was higher than other honey samples. Also Hakkâri region honey showed antimicrobial activity against 2 different bacterial species and Candida albicans.
As a result, the antimicrobial activities of bee products were tested against 5 Gram-negative, 7 Gram positive bacteria and 3 yeast species. The bee products which had been analyzed in this study showed high antimicrobial effect.

[P.16.379] Comparison of Commercial and Anatolian Bee Venom in Terms of Chemical Composition

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In this study, we compared the samples of bee venom, which were sold commercially and specially produced by Anatolian beekeepers for apitherapy and cosmetic purposes. For this purpose, the contents of the bee venom obtained by electroshock method were analyzed. In this study, 2 commercial bee venoms and 3 specially produced by Anatolian beekeepers bee venoms were used. In all samples, the % moisture content was determined by using the moisture analyzer, and the sugar profile analysis was carried out using% Mellitin, Apamine, Phospholipase A and HPLC-RID using HPLC-UV. The results were compared with the statistical method and it was concluded that commercial bee venom and Anatolian bee venom have different physico-chemical properties. Consequently, the standard quality criteria of bee venom to be used should be determined. The chemical content of bee venom should be evaluated according to the standard quality criteria.

[P.16.380] Sweeten the honey pot with quality analysis at affordable prices

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Nowadays beekeepers and honey traders are increasingly confronted with a wide range of different analyses from different testing laboratories. In addition, the worldwide trade means that there are many different requirements for import and export. You can quickly lose

track and spend too much money on unnecessary testing or even taking risk of unwanted properties of the product.
A short overview about common testing methods and existing quality systems is given. We want to show different strategies to streamline testing and reduce costs by utilizing a risk-based approach, pooling of samples, and taking advantage of analytical packages.
We aim to show you through illustrative examples that competitive laboratory analysis can simplify your process and lead to an increase of profit for your business.

[P.16.381] Comparison between invertase activities of raw honey and pasteurized honey

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Honey has a complex structure of different components and also contains enzymes which are responsible for its antibacterial properties. The most common enzymes in honey are diastase and invertase. The enzyme content of honey varies according to its botanical origin, region and season. In the past, diastase enzyme content was used to determine the quality of honey.
Invertase enzyme breaks the sucrose into fructose and glucose in honey and it is known to be more sensitive to heat than diastase enzyme. Therefore, the invertase content can be used as a marker to differentiate raw honey and heated honey.
In this study, 81 raw honey samples which had been supplied from beekeepers and 38 honey samples which had been purchased from different shops in Turkey were compared on the basis of their physicochemical properties including moisture, conductivity, diastase, proline, acidity, HMF and invertase. Standard methods were used for all analyses.
The mean values of invertase in raw pine, chestnut and flower honey were found as 210,9 U / kg, 180,6 U / kg and 186,1 U/kg, respectively. On the other hand, the average value of invertase in the honey which had been supplied from the market varies between 44.5 U/kg and 51.5 U/kg.
All in all, it is seen that invertase value is a better marker which reveals honey freshness, storage conditions and heat treatment rather than diastase and HMF value of honey. HMF and diastase values of honey remain within the legal limits in spite of heat treatment while a rapid decrease is observed in invertase value of honey.

[P.16.382] Global warming and HMF: are the Codex levels too low?

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Global warming is the long-term rise in average temperature of the Earth's climate system due to increasing greenhouse gases such as carbon dioxide, carbon monoxide, nitrous gases and methane from human induced activities and fossil fuel emissions. Although much discussion has been made about the effects of these rising temperatures on human living standards, warming oceans, loss of aquatic life, melting polar ice sheets and glaciers, little has been noted about the impact on honey quality while it is in the hive or under storage after harvest.
Currently hydromethylfurfural (HMF) is used as a quality measure to determine excess heat treatment for honey at the point of sale. Codex states that honey should contain no more than 40 mg/kg from temporal climates and no more than 80 mg/kg from tropical environments. But how realistic are these values with increasing mean summer temperatures and an influx of lower-economy (rural) beekeepers who may not have access to cold storage for their honey?
This talk will explore a range of HMF values from temporal and tropical honeys to show that if honey is to meet Codex requirements, then shelf life of freshly harvested honey may to be a little as 3 months before HMF levels exceed Codex standards. Depending on local temperatures and harvesting conditions, honey may already exceed these levels at the point of extraction. This talk is designed to provide thought towards a revision of the Codex HMF levels in honey and research required to determine food safety consequences of higher HMF levels in honey, especially where honey is stored in warm conditions (above 20 C) and consumed over several years.

[P.16.383] Production of spray dried honey powder and its application in food industry

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Honey is a popular sweet food with numerous health-promoting effects. However, its high viscosity and stickiness nature presents difficulties in transportation and consumption. Conversion of honey to its powdered form facilitates its application in food and drug industries. Spray drying is one of the most frequently dehydration techniques. It is a complex process which could be influenced by raw material characteristics, process variables and equipment design. The presence of low molecular weight ingredients in honey could form weighty sediments on the walls of drying chamber. Addition of carrier agents which have high glass transition temperature such as hydrocolloids into the inlet feed could decease the stickiness during spray drying. To avoid from economic loss and operating problems, the drying conditions including inlet temperature and carrier concentration have to be optimized. At this research, challenges in production of honey powder using spray drying technology were reviewed. Moreover, a new formulation for spray drying of honey using 40% carrier agent (maltodextrin + special sugar) was offered.

[P.16.384] Melissopalynological and physicochemical caracteristics of honeys produced in Norththwestern Algeria

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Objective / Purpose : Algeria is considered a contry traditionally consummer of honey. The lack of national legislation contribute to promote fraud and falsification. Research oriented toward assessment of the pollen spectra and physicochemical caracteristics of honeys according to the regions of their harvests allows to promote the local products on the market and may increase the commercial value of these products. This work contribute to the valorazation and knowledge of honeys characteristics from northwester of Algeria.
Material and Methods : The pollen spectrum study of 28 honeys collected during 2015 and 2016 harvest from different regions in northwest of Algeria were performed using a melissopalynological method (louveaux et al., 1978). In addition the physicochemical proprieties of these honeys including pH, moisture, electrical conductivity, diastase activity, hydroxymethylfurfural (HMF), color, were evaluated following standard methods. sugar profil were mesured by the NMR method (nuclear magnetic resonance).
Results : A total of 94 pollen types belonging from 46 families were identified. The highly represented families are Leguminosae and compositae. 19 samples were classified as monofloral and characterised by 13 dominant pollen types: Eucalyptus, citrus, zyziphus lotus, capparis, Genista, Punica granatum, Foeniculum vulgare, Apiacea, capparis, centaurea, Tamarix, Eriobotrya, melilotus. The presence of considered non nectarifarious species as olea europea is frequently hight being in some samples the principal type in the pollen spectrum of the honeys. Only 28% of the appellation presumed by beekeepers turned out to be correct. The overall analysis of honey moisture, pH, electrical conductivity, HMF, diastase activity, color, fructose, glucose, disaccharide were found to be 17.33%± 1.32, 3.99 ± 0.19, 0.512 mS/cm ±0.23, 21.45 mg/kg ±2.13, 8.56 (Schade units) ± 5.75, 82,39 (mm Pfund) ±31.55, 42.5 g/100 g, 31g/100 g, 5.75g/100 g respectively.
Conclusion : The pollen spectra of honey from northwest of algeria correspond to samples produced in semi arid areas of mediteranean vegetation. In general the samples were found to meet the requirements of the international honey standards.

[P.16.385] The Ecological Problems and Possible Solutions of Beekeeping in Hills and Terai of Chitwan, Nepal

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A general survey accomplished in hills and terai of Chitwan, Nepal in 2004 with an objective to investigate the ecological problems of bee keeping in chitwan and to recommend solutions for its improvement. The survey identifies predators, parasites, diseases, pesticide poisoning, and bee pasture as the key ecological problems in the Hills. Simultaneously, deforestation and pesticide poisoning are the concerned ecological problems in Terai. A very high majority of beekeepers in hills and more than half in Terai faced colony absconding problem in Chitwan. The general survey realized on the need of a special package of program to over come these problems, which include advanced apicultural research and educational activities for the commercialization of beekeeping and advance level crop pollination management in Nepal. For this a clear-cut beekeeping policy and guideline should be developed andshould be implemented.

[P.16.386] Physicochemical and Functional Properties of Propolis Added Ice Cream

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Propolis is a natural resinous substance that is formed by collecting bees from some parts of plants, buds and the secretions of plants. It has important functional properties such as strong antioxidant, antimicrobial, antitumor and antiinflamatory activites. For that reason, propolis has important potential for producing new functional foods.
Ice-cream is a widely used and popular milk product with its own taste, aroma and texture and it is a good nutriton source since its raw material is milk. In this study, it is aimed to provide a functional property to the ice cream by adding propolis in different proportions. In the freezing mix to be prepared, 6 groups of ice cream will be obtained which will contain control group and 0.1%, 0.2%, 0.3%, 0.4%, 0.5% propolis powder. Antioxidant , mineral matter; dry matter, titration acidity, viscosity, volume increase index, melting ratio, texture and sensory analysis have applied to samples of each group.
Significant difference at phsysicochemichal properties has not been observed between groups. Increase at antioxidant activity has detected directly proportional to propolis amount. Sensory acceptibility amoung groups was similar except 0,4% and 0,5% propolis added groups.

[P.16.387] Authenticity of Manuka Honey - HAHSUS versus Test Defined by the Ministry for Primary Industries (MPI)

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Manuka honey is traded at very high market prices due to its anti-micro-bacterial properties. Since the demand far surpasses the supply, manuka honey is a typical object for food fraud. To authenticate manuka honey, the Ministry for Primary Industries (MPI) has decided that a combination of five attributes (4 chemicals, 1 DNA marker from manuka pollen) is to be required for the differentiation of manuka

honey from other honey types and for the identification of monofloral and multifloral manuka honey. However, said attributes are not suitable for protecting consumers from manuka honey fraud. The listed substances can be purchased at a reasonable price from the commercial chemical trade, and the positive DNA test required can be obtained already with a mere 10-percent amount of manuka. In contrast, the HAHSUS model (Honey Authentication by HS-SPME-GC/MS and UHPLC-MS/MS combined with Statistics) developed by the work group offers distinct advantages: via TLC, the components 3,6,7-trimethyl-2,4(1H,3H)-pteridinedione and 6,7-dimethyl-2,4(1H,3H)-pteridinedione are initially tested for [1,2]. Subsequently, the profiles of the volatile substances and the non-volatile substances are recorded. Aside of the marker substances, the profiles are tested for typical peak sequences. By recording relative diagrams using marker substances unavailable from the commercial chemical trade, the percentile amount of manuka honey can even be determined for manuka/kanuka mixtures [3].
Thus, compared to the MPI attributes [4], the HAHSUS model is more suitable for the avoidance of food fraud.

[P.16.388] Metabolomic approach for the identification of honey adulteration with sugar following bee feeding: preliminary study

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Honey is a natural product with an excellent nutritional value. The limited availability and high price of honey contribute to encourage the illegal practice of adulterating honey. The most frequently used methods for honey adulteration are the direct addition of sugar syrups and the overfeeding of bees with syrup during the main honey-producing season. As in other food products, honey adulteration has evolved over time from the simple addition of sugar and water, to the use of feedings whose chemical composition reproduces the sugar composition and ratios of honey. It is therefore needed to develop more reliable methods to detect adulteration. The purpose of our work is an alternative approach to the identification of potential markers to detect honey adulteration with feed produced from beet. We performed an untargeted metabolomic study using liquid chromatography coupled to high resolution mass spectrometry (HRMS). Metabolomic studies are based on electrospray ionization (ESI). The advantages of this relatively soft ionization technique include minimal fragmentation, an excellent quantitative analysis and a good analytical sensitivity. Ionization is performed both in positive and negative mode, allowing the detection of two sets of complementary analytes. By means of HRMS Q-Exactive (Thermo Fisher Scientific), the detection in positive and negative ionic mode can be performed simultaneously in a single run, thus reducing both the analysis time and the injection variability. The acquisition of the high resolution mass spectrum (70,000 FWHM) of the metabolomic profile of each sample was performed in the range of 70 to 1000 m/z. The study included the processing of data derived from the metabolomic analysis (software Thermo Scientific SIEVETM), the verification of the selected masses in fortified honey samples and samples derived from feeding sugar to colonies, and the application of the method to real samples of honey. Six molecules were identified and in particular, one that seems to be a promising selective marker for complementary feed originated from beet. The promising marker was detectable down to 5% adulteration level. A possible identification of these molecules is in progress. This work represents the first study on honey adulteration with an untargeted metabolic approach.

[P.16.389] Authentication of honeydew honeys by analyzing non-volatile components

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Honeydew excreted by aphids feeding on conifers in Germany and Central Europe is an important food source for bees in late summer. The honeys produced from it are of a dark color and have an especially spicy, malty taste. Many consumers like these properties and accept the higher prices.
So far, honeydew honeys are distinguished mostly by their sensory properties. The classical microscopy pollen analysis failed due to the missing pollen. In order to protect the quality and the authenticity of these rare, expensive honeys, a project called “BooglH” (botanical, zoological, and geographical identification of honeydew honey) was launched and funded in Germany by The Federal Ministry of Food and Agriculture (BMEL). The aim of the German “BooglH” project is to provide an accurate definition of honeydew honeys by means of objective chemical-analytical methods in order to promote their marketing and to discover food fraud.
For this purpose, our group has developed a multimethod for the determination of various non-volatile honey components. The detection of individual honey substances was carried out by SPE-(U)HPLC-PDA-MS/MS. The chromatographic profiles of a total of 42 authentic fir, spruce, and pine honeys were compared, and marker substances were identified by using chemo-metrics. Employing a discriminant analysis based on the best specific markers, a differentiation of the botanical origin of honeydew honeys was achieved. Afterwards, it was possible to classify more than 20 commercial honeydew honey samples from the market. Consequently, honeydew honeys can objectively be tested for their purity for the first time.
Acknowledgement:
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[P.16.390] Development of a Rapid Honey Screening Immunoassay for residues of Nitrofurans AOZ, AMOZ, AHD, SEM

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Background: Nitrofurans are a class of broad spectrum antibiotics which have been banned for use in food producing animals in the European Union and USA as a result of their potential carcinogenic effects following human consumption. Monitoring of their illegal use in food produce is vital to ensure food safety but is complicated by the short in vivo half-life of the parent compounds. However, tissue-bound metabolites of these drugs, including AMOZ, AOZ, AHD and SEM, are stable for several weeks after antibiotic use and are more reliable markers for detection of illegal Nitrofuran use.
Method: Four easy-to-use, ELISA kits have been developed to facilitate detection and quantification of each of the Nitrofuran metabolites AMOZ, AOZ, AHD and SEM in honey samples. Honey can be prepared, following a single sample preparation protocol, for analysis on all four Nitrofuran kits. Briefly, 1g honey undergoes acid hydrolysis and simultaneous derivatisation by 2-Nitrobenzaldehyde at 60 degrees Celsius for 90 minutes. Solvent extraction is then utilised to yield the Nitrofuran metabolites for quantification by ELISA.
Results: Excellent intra-assay precision has been demonstrated across all four Nitrofuran kits. At concentrations of 0.09ppb and 0.2ppb, CVs for AHD were 4.2% and 4.3%, respectively. CVs for AMOZ were 5.0% and 3.9% at concentrations of 0.08ppb and 0.25ppb, respectively. For SEM, CVs were 2.7% and 4.7% at concentrations of 0ppb and 0.1ppb, respectively. At concentrations of 0ppb and 0.125ppb, CVs were 3.6% and 3.4% for AOZ, respectively. Sensitivities, expressed as the half-maximal inhibitory concentration (IC50), were 0.09ppb for AHD, 0.21ppb for AMOZ, 0.21ppb for SEM, and 0.20ppb for AOZ.
Conclusions: The four Nitrofuran ELISA kits developed exhibit good sensitivity and excellent intra-assay precision. All four kits have a short assay duration of 45 minutes and, importantly, a single sample preparation is compatible with all four kits, significantly reducing labour and overall costs. Additionally, in comparison to other commercially available Nitrofuran ELISAs, these new kits display a number of advantages including a sample preparation time which is 30 minutes shorter than that currently on the market, and lower solvent volumes which make the sample preparation more cost effective.

[P.16.391] Volatile compounds for differentiation of honeydew honeys using HS-SPME-GC/MS

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The commercial interest in honeydew honeys has been increasing due to their higher antibacterial and antioxidant properties and because their flavor is spicier and maltier than that of blossom honeys. However, an accurate definition of honeydew honeys using objective chemical-analytical methods is necessary.
For this purpose, the volatile components of the honeydew honeys were investigated in addition to the non-volatile ingredients as part of the "BooglH" project. Headspace solid phase microextraction (HS-SPME) coupled with GC/MS is the most commonly used technique for the extraction and analysis of the volatile fractions of honeys. With this analytical method, a variety of aroma compounds have already been reported as markers in blossom honeys by Oelschlaegel [1]. In the case of honeydew honeys, a few markers are known but only for some types. German forest honeys have rarely been part of these studies. Therefore, in this study, a total of 42 authentic pure honeydew honeys were studied – 14 silver fir honeys (Abies alba), 17 Norway spruce honeys (Picea abies) from Germany, and 11 pine honeys (Pinus species) from Turkey. Six lime honeys (Tilia species) and six chestnut (castanea sativa) honeys from Germany and Europe, checked by pollen analysis, were also considered in the study as they often occur in Germany in mixtures with forest honey.
The aroma profiles of all 54 honey samples were investigated by a modified HS-SPME-GC/MS method according to Oelschlaegel. In total, 38 volatile compounds were extracted for peak integration. Compared to many blossom honeys, the honeydew honeys had low-intensity flavor profiles. Nevertheless, it was possible to establish volatile marker compounds for honeydew honeys by means of chemo-metrics. A differentiation of the botanical origin of honeydew honeys was achieved by principal component analysis.
Literature
[1] Oelschlaegel S., Characterization of unifloral honeys using HS-SPME-GC/MS, 42nd APIMONDIA 2011, Argentina.
Acknowledgement:
The BooglH project (No. 2816500314) is supported by funds of the Federal Ministry of Food and Agriculture (BMEL) based on a decision of the Parliament of the Federal Republic of Germany via the Federal Office for Agriculture and Food (BLE) under the innovation support program.

[P.16.392] Traceability of potential enterotoxigenic Bacillus cereus from bee-pollen samples from Argentina at different sampling throughout the production process

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Bee-pollen is the result of the agglutination of pollen grains collected from flowers and mixed with nectar and salivary secretions by honeybees. Bee-pollen is a functional food sold for human and animal consumption but also is a favorable microhabitat for many spore-forming bacteria. Among them, Bacillus cereus is a ubiquitous Gram-positive spore-forming bacterium found on soil, plants, and enteric tracts of insects; these niches include honey and pollen. B. cereus can produce several toxins and other virulence factors causing an emetic or diarrheal syndrome after ingestion. Beekeepers from South West of Buenos Aires Province, Argentina collect bee-pollen from hives by using traps and transport it to the processing unit for freezing (-10 °C for two months), dehydration (40°C for 24 h), cleaning, and packaging. This work aimed to study the traceability of potential enterotoxic Bacillus cereus based on colony counts, rep-fingerprinting and toxigenic profiles at four sampling points of the production process. Thirty-six bee-pollen samples from three beekeepers of SW area were analyzed between February and June 2016 by testing three samples from each beekeeper at each sampling point. Forty-seven isolates of B. cereus obtained in PEMBA yielded 24 different patterns by using BOX and ERIC primers. Isolates were also evaluated for the presence of sequences associated with virulence genes by PCR. Genes encoding for hemolysin BL (hblA, hblB, hblC, hblD), enterotoxin-T (bceT), cytotoxin K (cytK), non-hemolytic complex (nheA, nheB, nheC), sphingomyelinase (sph) and cereulide (ces) were studied. A positive correlation was observed between rep-fingerprint patterns and the enterotoxigenic profiles obtained. Colony counts on PEMBA revealed that B. cereus incidence significantly increased from collection to dehydration and slightly decreased in the final step of cleaning. Also, cross-contamination occurred as shown by differences in fingerprint patterns after freezing, dehydration and cleaning steps compared to the initial collection step.

[P.16.393] Full Profile for Honey Testing with both Multiplexing Technology for Drug Contaminant Screening and Quality Testing for the Detection of Adulterated Honey

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Introduction. Antibiotic drug residues in honey pose a potential risk to human health though they are used worldwide in apiculture to treat or prevent bacterial diseases. Biochip array technology (BAT) allows the multiplex screening of antibiotics from a single honey sample.
Fructose and glucose are the main sugars present in honey. Hydroxymethylfurfural (HMF) is produced by acid-catalyzed dehydration of sugars. Sugar content of honey has been considered a measure of the purity and important in quality control of honey. Elevated concentrations of HMF provide an indication of overheating, poor storage conditions, possible adulteration with other sugars or syrups and/or higher age of the honey. The development of analytical methods which monitor the compliance to quality specification, facilitates fraud prevention and the protection of authentic honeys.
This study reports the analytical performance of two biochip arrays for the multi-drug detection and two assay kits one for the determination of D-glucose and D-fructose and the other for the determination of HMF.
Methods. Multiplex screening of antibiotics: simultaneous competitive chemiluminescent immunoassays defining discrete test sites on the biochip surface and applied to the biochip analyser Evidence Investigator were employed. Antimicrobial Array II Plus for the simultaneous detection of ceftiofur, quinolones, streptomycin, tetracyclines, thiamphenicol and tylosin. Antimicrobial Array V for the simultaneous detection of chloramphenicol and nitroimidazoles
Assay kit for D-glucose and D-fructose: the UV method is based on enzymatic reactions Assay kit for HMF: Differential UV-photometry method with and without sodium bisulphite-reduction of HMF. These assays were applied to the analyser RX misano.
Results. Antimicrobial array II Plus: Limit of Detection (LOD) from 1 ppb (thiamphenicol, tylosin) to 4.5ppb (tetracycline), intra-assay precision (n=20) < 9.5%. Antimicrobial Array V: LOD 0.1ppb (chloramphenicol) and 0.9ppb (metronidazole), intra-assay precision (n=20) < 8.5%.
D-glucose, D-fructose and D-glucose + D-fructose: sensitivity and linearity values were 0.3g/L and 7.5g/L respectively, intra-assay precision (n=20) <4%. HMF: sensitivity and linearity values were 2mg/kg and 100mg/kg respectively, intra-assay precision (n=20) <20%.
Conclusion. Results indicate that the biochip arrays for multiplex detection of drug contaminants and the assay kits for the detection of sugars and HMF are reliable analytical tools for honey testing.

[P.16.394] Development of a Rapid Honey Screening Immunoassay for residues of Metronidazole and Dimetridazole

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Background: Nitroimidazoles such as Metronidazole and Dimetridazole are a family of antimicrobial agents with antibacterial and antiprotozoal activities. They are of particular interest in the honeybee industry as a cheaper alternative to other drugs known to prevent and control the parasitic disease Nosemosis/Nosema. However, in the European Union (EU), their use has been prohibited in all food-producing species (Group A6 of Annex I of Directive 96/23). Despite this, contamination of honey with Nitroimidazole residues has become a significant issue in the EU, with Metronidazole residues in particular being detected in honey imported from non-EU countries. In response, to control the integrity of honey in the EU, the Community Reference Laboratories (CRL) have released a Guidance Paper that requires detection methods capable of detecting a minimum of 3µg/kg (3ppb) Nitroimidazoles in honey.
Methods: A sensitive, easy-to-use ELISA kit has been developed to facilitate the detection and quantification of Metronidazole in honey. To use this kit, samples can be prepared from 3g honey following a simple solvent extraction protocol which involves addition of ethyl acetate to the sample, followed by its evaporation, and subsequent reconstitution of the sample for application to the ELISA plate.
Results: The calibration range of the assay is 0.625 – 20ppb. Excellent intra-assay precision has been demonstrated at concentrations of

1.25ppb (CV 2.5%) and 5ppb (CV 2.5%), with cross-reactivities of 100% for Metronidazole and >100% for Dimetridazole. Analysis using honey samples has demonstrated a CCbeta (detection capability) of 3ppb, with recoveries of 90.8 – 122.0%.
Conclusion: The Metronidazole ELISA kit exhibits excellent intra-assay precision and sensitivity in relation to the minimum requirements set by the CRL. With high cross-reactivities for Metronidazole and Dimetridazole, this kit can be used to detect contamination of honey with both Nitroimidazoles in as little as 55 minutes. Combined with a simple, short sample preparation method, this kit represents a low-cost option for reliable detection of Nitroimidazole contamination in honey both produced in and imported to the EU and other countries.

[P.16.395] Development of a rapid multi-analyte lateral flow tests to detect antibiotics in honey

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Cheap, rapid, cost effective multi-target antibiotic residue screening is currently required in the honey industry. Simultaneous detection of antibiotic residues would be a major advantage for the honey industry reducing time and cost of analysis of products for positive release to market and avoiding expensive recalls thereafter. The objective of the EU-funded Eurostars project LOGIC (www.logic-multiplex.com) is to develop a prototype kit for multi-analyte antibiotic screening in several food matrices that is suitable for use by the food industry.
At BioSensing & Diagnostics, multi-analyte diagnostic platforms such as the lateral flow microarray immunoassay are being developed. These test formats are rapid Point-of-Care lateral flow microarrays and can consist of as many as 25 assay spots; with sample detection possible within a few minutes. Our test will enable screening of raw honey samples to take place at point of origin/receipt and ensure processes such as blending are not delayed. This test will give honey processors a higher degree of control of all raw honey entering their plants.
Current honey sample preparations for some antibiotic targets require long laborious derivatisation incubation while our test aims to reduce this incubation/labour time and use green “environmental friendly” solutions where possible. It is anticipated that antibiotic concentrations in honey samples for this test will be read using a new and innovative real-time video reader, which yields real-time and quantitative data. The standalone reader can be controlled wirelessly by a smartphone and can also be used to transfer data to receivers such as a GP, a (pre-) clinical laboratory or directly to a patient’s smartphone.
Initial results for the development of this user-friendly test for the detection of antibiotics of the 4 drug families (nitrofurans, tetracyclines, nitroimidazoles and chloramphenicol) will be shown at the conference.

[P.16.396] Actual requirements of the global honey market concerning control of antimicrobials residues

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Study of the dynamics of honey export from Ukraine shows its steady growth over last ten years. During this period of time, Ukraine has entered leading positions among countries exporting honey. Nevertheless, certain problems remain due to the cases of detection of antimicrobial drugs residues prohibited by national and international law, which makes the control at all stages of export honey lots formation to be obligatory. Considering the requirements of export contracts regarding methods sensitivity for antimicrobials determination, over past five years the demands to detection limits and the actual list of substances to be controlled, have changed. For antimicrobials listed in Table 2 of EC Regulation 37/2010, they are 0.05, 0.1 and 0.2 ppb for chloramphenicol, nitroimidazoles and nitrofurans metabolites, respectively. In addition, in recent years, we have discovered first cases of dextramycin (chloramphenicol enantiomer) residues detection in honey, which presence makes immune methods ineffective for honey incoming control. Previously the list of most common antimicrobial honey contaminants was headed by chloramphenicol, tetracyclines, sulfonamides, streptomycin and nitrofurans metabolites, while now sulfathiazol, metronidazole, dextramycin, AOZ, SEM and erythromycin detection is more frequent.
Discovering of new threats requires sufficient changes in the technology of honey control in industrial and reference laboratories. Permanently we analyze main trends affecting the cost of testing of obligatory safety parameters in export honey batches. According to our conclusions, the most effective solution for honey control is UPLC-MS/MS technique for simultaneous detection of representatives of several widespread groups of antimicrobials. Still, the use of high-cost UPLC-MS/MS method in the laboratories of exporting companies requires significant investment, which influences the formation of market prices for honey. It is possible to reduce the cost of required researches on honey export lots by simultaneously detecting all potentially dangerous analytes in a single sample during short analysis procedure without complicated sample preparation. The implementation of these conditions is an actual challenge for analysts and requires correct problem statement and the choice of ways to solve it. The approaches we introduced have allowed the leading Ukrainian companies to adopt UPLC-MS/MS methods in their control laboratories during last four years.

[P.16.397] Bee pollen nutritional value and microbiological stability: influence of preservation techniques

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Bee pollen is often considered a highly nutritive foodstuff. After its collection, the bee pollen can present moisture contents ranging from 18% to 25%, depending on the gathering season and trapping technique [1]. These moisture contents, combined with the high nutritional value of bee pollen, provide conditions highly favorable to microorganism growth and undesirable pollen fermentation. Moreover, when not preserved adequately, its nutritional value can be quickly reduced, due to Maillard reactions [2]. Thus, after collection it is necessary to reduce the bee pollen moisture content in order to maintain its overall nutritional quality and microbiological safety. The main objective of this work is to provide an insight regarding the potential influence of the applied preservation techniques on the nutritional value and microbiological quality of bee pollen.
The bee pollen, which was collected in beehives located in the northeastern Portuguese region of Bragança, had a moisture content of 13.8%. Subsequently, the fresh pollen was submitted to various preservation techniques, namely oven drying at three distinct temperatures (35ºC, 40ºC, and 45ºC) and freeze-drying. The pollen samples dried at 35ºC, 40ºC, and 45ºC presented moisture contents of 9.6%, 9.8%, and 10.1%, while the freeze-dried sample had a moisture content of 5.8%. The nutritional value of the preserved bee pollen was assessed throughout time during a period of 6 months, through the determination of the moisture, ash, protein, crude fat, and carbohydrate contents. The effect on the microbiological quality was also analysed, and included the parameters: total viable counts (aerobic mesophiles), lactic acid bacteria, yeasts and molds. In general, the different treatments showed no significant immediate impact on the microbiological loads of bee pollen, but changes were observed after one months of storage, mostly in total viable counts and lactic acid bacteria.
References
[1] Casaca, J.D. (2010). Manual de produção pólen e própolis. Federação Nacional Apicultores de Portugal (Eds.), Lisboa.
[2] Almeida-Muradian, L.B.; Pamplona, L.C.; Coimbra, S.; Barth, O.M. (2005). Chemical composition and botanical evaluation of dried bee pollen pellets. J. Food Compos. Anal., 18, 105–111.
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[P.16.398] Phytochemical, physicochemical and pollen characterization of the organic honeys of the Moroccan Atlas

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Moroccan atlas organic honey is a natural product made from medicinal plants. Its richness in bioactive compounds gives it nutritional and therapeutic properties.
This work aims at characterizing the quality of sixty-four samples of mountain organic honey collected from different Middle Atlas stations in Morocco. The characterization of honey is carried out by analyzing the physicochemical properties, antioxidant capacity and the botanical origin by the pollen analysis. The microbiological quality of the different honey samples was also determined.
The physicochemical parameters were evaluated by analyzing 12 parameters recommended by the European quality control and food safety legislation. In addition, tests of antioxidant activity were performed to evaluate the antioxidant content of organic honey samples.
The results of the physicochemical parameters confirm that all our organic honey samples respect the high-quality control standards. Preliminary analyzes of honey samples studied have shown an undeniable richness in polyphenolic compounds including syringic acid and chlorogenic acid.
Microbiological analyzes confirm that these samples have an acceptable microbiological profile; none of the analyzed samples contains microorganisms having an impact neither on the organoleptic quality nor on the health of the consumer. Finally, the results of the botanical analysis showed new non-listed pollen from endemic medicinal plants as well as the results show the authenticity of the biological honey collected and the information pronounced by the beekeepers.

[P.16.399] The influence of the application of new regulations on the production of honey in the republic of Serbia

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The main objective of this study was to review the effects of the application of the new Regulation on the declaration, labeling and advertising of foods to honey producers in the Republic of Serbia, through the evaluation of quality, sensory properties and labeling of eighty different honey samples collected from markets in Serbia and verifying their legal compliance. Physicochemical properties, sensory and labeling

of three honey types acacia, blossom and honeydew were studied. In order to prevent the deceitfulness of end-users, especially in terms of: characteristic food properties and contents, ascribe attributes and properties that does not possess, indicate the specific properties, if similar food has the same characteristics, all of these requirements are also refer to honey, and honey producers must respect them. As a single ingredient unprocessed food, honey does not require to be labelled with a nutritional declaration. If a nutrition declaration is provided on honey it must be accurate and presented together in a clear format using the correct units of measurement as well as the correct order of presentation. Methods established by the International Honey Commission (IHC) as well as AOAC official method were used. Sensory analysis was carried out using method of quantitative descriptive analysis and the color, taste, smell, viscosity of honey and labeling were evaluated.
This paper gives an overview of the current state of the application of Regulation requirements in regard to the quality of honey, sensory and the correct use of the products declaration. It was concluded that the quality and sensory properties were not significantly changed, but only 26% were correctly declared.

[P.16.400] Quality Criteria of Samar and Sider Honey Produced Under GCC Conditions

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Samar (Acacia spp.) and Sider (Ziziphus spp.) honey have distinctive properties associated with strong taste and odor. These lead to nonconformity of Samar and Sider honey with current standard limits of GCC and UAE standards (UAE.S GSO 147:2008) especially for acidity and electrical conductivity values. Study the physicochemical characteristics of Samar and Sider honey that differ according to its botanical origin and geographical location can be the starting point on the preparation, and standardization criteria of locally produced honey in the GCC region. Various chemical and physical parameters are used to characterize honey quality. These criteria include moisture content, water activity, acidity, HMF, fructose %, glucose %, sucrose %, F/G ratio, diastase activity, and color. Total of seventy-one honey samples was collected directly from beekeepers from different countries in the region. Twenty-three samples of Samar honey from Yemen (3) and Jordan (3), UAE (5), KSA (12), and forty-eight samples of Sider honey collected from Kuwait (3), Yemen (3), Jordan (3), KSA (18), and UAE (21). Samar honey normally shows a high level of total acidity (60-80 mEq/Kg) that does not correspond to UAE standards (not more than 50 mEq/kg) compared with Sider honey (0-40 mEq/kg). Samar and Sider honey have a similar range of the other chemical parameter values such as fructose (25-45%), glucose (20-35%), moisture content (12-18%) and total reducing sugar (50-75%). There is a significant difference in color between both types associated with the strength of flavor. Samar honey has dark color varied from brown to very dark brown color and has range of L* values between 3.87 to 17.95, a* values between 1.55 to 12.74 and b* values between 4.16 to 52.16 compared with light to dark golden color of Sider honey that has ranged between 6.22 to 11.83, -3.40 to 4.94, 6.00 to 13.27 respectively.

[P.16.401] Physical and chemical quality of honey from hives feeded with suplementary feeding

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The aim of this study was to evaluate the influence of honey bees feeding with different sugar pastes all manufacturated in Zukán S.L (Murcia, Spain): sugar paste (Apipasta®), sugar paste + vitamins + free aminacids (Apipasta Vitamina®) and sugar paste + vitamins + crude protein (3%) (Apipasta Plus®), in order to act as a pollen substitute, to examine the effect on the quality parameters and mineral composition of honey.
The experimental study was carried out in the beekeeping unit of University of Murcia, Faculty of Veterinary. Twenty Eight colonies were distributed randomly into four groups being bees feeding from December 2017 to April 2018. Once the honeys were obtained, the quality parameters were analyzed, according to those established in the European Honey Directive. The sugar profile, Hydroxymethylfurfural, Diastatic Activity, Moisture, Color, Electrical Conductivity, Free Acidity, and mineral composition was determined.
All honey samples analyzed from the different groups were in accordance with the criteria described in Directive 2001/110/CE. However, significant differences ($p<0.05$) were detected in Color, Free Acidity, Diastase Activity, Hydroximethylfurfural, sugar profile and Conductivity between all honeys.
In terms of mineral content, significant differences ($p<0.05$) were found. The honeys from hives feeded with Apipasta Plus® and control group had higher values for Na, Mg, P, K, Ca, Mn, Fe, Cu, Zn than hives feeded with Apipasta Vitamina® and Apipasta®.

[P.16.402] Rapid, easy to use and cost effective method of analysing glucose/fructose in honey samples

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Honey is used for nutritional, medicinal and industrial purposes and is therefore a very important global commodity. While sugar is 100% sucrose, honey is composed primarily of the simple sugars glucose and fructose (monosaccharides) and a further 17% to 20% of water. Fructose is slightly sweeter than glucose and is the predominant sugar in most honeys, making the honey taste slightly sweeter than sugar. Of course these proportions may vary depending on the source of the nectar. Differences in climatic conditions and vegetation are also important factors that can affect the various properties of honey and can act as indicators of origin source of particular honey samples.
These 2 carbohydrates are responsible for some of the key functional properties in honey, to include the ability to hold moisture and extend shelf-life, its microwave reactivity and its ability to promote colour and flavour. Another key feature of honey carbohydrate composition is crystallization. The fructose/glucose and glucose/water ratios are used to help predict the tendency of honey to crystallize. Honeys with a lower glucose/water ratio generally will not crystallize readily.
Although no limits have been fixed for individual Glucose/Fructose values in honey, their sum (Fructose + glucose) has been fixed at a value of more than or equal to 60 g/100 g as one of the requirements of the international standard for honey established by Codex Alimentarius Commission. Due to this regulation there is a need for the frequent analysis of these sugars in honey. In good quality honey the fructose content should generally exceed that of glucose.
This simple to use test format enables honey producers to analyse their honey samples in real-time, without the need for expensive laboratory techniques, simplifying the method of analysis. In essence, the honey sample is diluted along with glucose and fructose specific reactive reagents. Following a simple step by step procedure these diluted reagents are applied to a dedicated Biorex Diagnostics benchtop biochemistry analyser. Results can be obtained in <10minutes and achieve precision of <10% along with excellent linearity, recovery and sensitivity.

[P.16.403] Determination of quality Iranian royal jelly by physicochemical properties

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Royal jelly (RJ) is generally considered as the main reason of the significant functional and morphological differences between queen and worker bees. The unique and chemically most interesting specifications of RJ is its fatty acids. RJ fatty acids are short-chained, 8-10 carbon free acids that are usually either dicarboxylic acids or hydroxy fatty acids. The main fatty acid in RJ is 10-hydroxy-2-deconic acid (10-HDA). [1-2]
In this study, we investigated and compared the physicochemical properties of RJ samples from diverse region of Iran, different seasons and bee race. Samples were analyzed for moisture, protein, total lipid and 10 HDA according to Iran's National Standards methods [3] and international standard [4]. The results obtained showed that moisture, protein, total lipid in all samples were in accordance with ISO Standard limits.
10-HDA was determination of quality in RJ samples by HPLC and result compared. The eluent was MeOH-H2O (55:45 v/v with pH=3.5). The wavelength and flow-rate were adjusted at 215 nm and 1 ml/min respectively. The external calibration method was used for determination of 10-HDA in samples. The minimum recovery determined found to be 98%. The results show that amounts of 10-HDA in Iran RJ samples was between 1.4-3.9% and the amount of this fatty acid in Pure RJ varies depending on the characteristics of the bee, origin of the jelly and different seasons. The aim of this study was determination of quality in royal Jelly and compared quality different RJ samples from diverse region of Iran in different seasons and bee race.
Keywords: Royal jelly, 10 HAD, HPLC

Reference

[1]. Honary S., Ebrahimi P. (2004). Determination of 10-hydoxy -2-decenoic acid in pure Iranian royal jelly by HPLC method. Iranian J. Pharm. Res. 259, 2: 86-86.
[2] Honary S., Ebrahimi P., Changizi Sh; Nozadi Z. (2004). Purification and identification of 10-hydroxy-2-decenoic acid from pure royal jelly. Iranian J. Pharm. Res. 260, 2: 86-86.
[3] Iran's National Standard No.18385. Royal jelly- Specifications. 1st. Edition, Apr. 2014.
[4] ISO No.12824:2016 Royal jelly-Specifications.

[P.16.404] Effect of heating on HMF and Daistase activity

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Honey is usually subjected to filtration and heating for bottling before commercialization.
However, there is no standard procedure available for thermal treatment on honey. Honey is thermally heated at various temperature and duration based on individual experience to prolong the shelf life of honey in the market. The heating methods might decrease the biochemical components such as nutrients, enzymatic activities and vitamins to certain extent. In addition to water reduction, thermal treatment on sugar rich honey usually accompanied by the formation of 5-hydroxymethylfurfural (HMF).
The short period of heating time was found to degrade nutrients, enzymatic activities and water soluble vitamins in honey. The degradation of protein and enzyme via proteolytic digestion had attributed to the increase of free amino acids in honey.

[P.16.405] Review on the Effect of Processing Temperature and Time duration on Commercial Honey Quality Parameters

Y. Eshete
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Raw honey after harvesting is usually strained and filtered to remove suspended materials including pollen, proplisand bee wax prior to heating for commercial processing. Heating honey is to facilitate filtering and bottling of honey by reducing the viscosity. In commercial processing plant, honey is usually heated in order to purify, filter,facilitate packaging, to inhibit microorganism growth, to reduce the moisture content at the standard level and to delay crystallization process. Even though heating is of great important in commercial honey processing, there is a limited information as to the required Temperature and time to heat for most honey types and regions. The optimal heating conditions are mainly relied on the geographical and botanical origins of honey. Uncontrolled temperature can be detrimental to the quality of honey and to its biological and bio active chemical properties. In Ethiopia, commercial honey processing industries can perform their processes in a different ways depending on various factors, including economic, technological and technique reasons. Honey has a probability to loss the natural quality when it goes through uncontrolled thermal processing. The loss of the natural quality is due to the decomposition of vitamins, destruction of the integrity of the enzymes and the development HFM content.HMF content and Enzymatic activities are the recognized honey quality parameters to penetrate and sustain in the international market. As heating is of great important on commercial honey processing industry, it needs to have standard guideline for the use of optimum heating temperature and time duration.

[P.16.406] Significant factors in concerned with a successful requeening

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Requeening hives is one of the most challenging aspects of beekeeping; it can either lead to a failure or a successful establishment of strong colonies. The important considerations and challenges include choosing the right time, finding the existing queens in highly populated hives, accessing (buying or/and rearing) high quality queens, acceptance of the new queens, as well as the economic factors, especially in commercial bee operations.
In this presentation, requeening at the beginning and at the end of the season is compared, to find a suitable time frame (long and short beekeeping seasons, that is geographical locations, had a great impact on choosing the right time frame) and a method that could bring a successful result.
Beginning of the season is most favored by beekeepers, as it is easy to find the existing queens because hives have a low brood production and a lower population; due to a sufficient flow of pollen/nectar, the colonies are well nourished and their acceptance rate of new queens is high. The challenge, however, is finding high quality queens, because queen production has not begun yet, so queens have to be imported from warm climate areas.
End of the season is the most challenging time, as the insufficient flow of nectar/pollen reduces nutrition among the colonies. This combined with a high level of pheromone emitted from larvae, induce bees to kill the new queens, also hives are highly populated which makes it difficult to find the existing queens. Additionally, this is the busiest time of the year for beekeepers, and requeening might not be the priority.
In locations where there is a 10-month beekeeping activity, we have to consider 30% to 50% of queen loss, for commercial beekeeping operations. The recommendation is to start requeening in mid-season, right after the first pollination, and to designate 15% to 20% of the operations to queen production and increase to maintain the size of the operations.

[P.16.407] A Queen for All Seasons: Trans-regional Survivor Stock & Longevity-based Breeding Program, Reflection of Living Laboratory Case Studies from Lake Superior to the Rio Grande to the Pacific

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Longevity of queens used in commercial/professional apicultural endeavors and hobbyist applications has continued to diminish as a result of multiple variables and circumstances. Though no one specific cause of such challenges has been deduced, there exist multiple stock lines and cross-stocks of diverse lineage/genetics that observably and measurably endure. The quest for such endurance in queens has fascinated producers across the globe. Queen bees who have been able to cope with today's deadliest of pests, Varroa destructor- vectors of pathogens are highly sought after by bee producers, diverse demographics of beekeepers, and researchers. There are queen breeders who have embraced the search to find, preserve, and breed these quality lineages which demonstrate longevity.
Zia Queenbees Farm & Field Institute- nestled at the kiss of the forests, where Santa Fe, Carson, and Pecos National Forests converge, on The High Road to Taos- is one such small-scale queen breeding operation. Located in the crenulated topography of the southern Rocky Mountains of northern New Mexico, ZQB utilizes the extreme landscapes of the Land of Enchantment- from desert to tundra and where the desert and the plains meet the mountains- to find, monitor and propagate those bees and queens which have demonstrated Overall Lifetime Merit (OLT). ZQB has dedicated its efforts to finding adaptable stock lines and cross-stocks that are able to perform well in multiple conditions, topographies, and under varied management styles by collaborating with beekeepers across the USA (MI, FL, CO, VT, OR, PA, NC, CA, HI, ID, WA). Despite the fragility of being a first generation, landless farming enterprise, ZQB has helped to nurture strain diversification of the genetic pool; and to support honeybees chosen by beekeepers for beekeepers through a Father Time Tested- Mother Nature Approved paradigm.
Longevity is a heritable, umbrella trait established over time sculpted by nature's dynamic interface. Selection of mother and drone breeders is based on a two-year thrive and survive regiment. Those that demonstrate productivity and additional characteristics of hygienics, pest/disease resistance, gentleness, and overwintering ability are eligible breeding contenders after successful completion through minimum two winters (18 months) with no commercial miticides.

[P.16.408] Standardization of sperm motility assessment in the honey bee drone

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Sperm motility is an essential characteristic of sperm quality. The method used to assess sperm motility in drones has mainly been based on the use of slide coverslips. The aim of the present work is to contribute to the standardization of honey bee drone sperm motility assessment by studying the effect of the viewing chamber on motility results. A total of 30 mature drones, 10 males from 3 colonies, were sampled individually. Semen was diluted in Kiev buffer before evaluation. We compared the effect of three different sample viewing devices: slide coverslips (SC), Makler (MK) and Leja10 (L10) chambers. Semen was placed in the viewing chamber and live video pictures were recorded. Sperm were classified subjectively as: motile sperm, if they presented any type of active movement, progressive sperm if the sperm head showed displacement and circular sperm if the sperm head and tail overlapped. Significant differences ($P < 0.001$) were observed for all parameters studied. A lower proportion of motile sperm was observed in SC chambers than in IS10 chamber, with mean differences of 18.0%. The percentage of progressive sperm was much lower in drop-loaded (SC and MK) compared to capillary-loaded chamber (IS10), while the percentage of circular sperm showed a contrasting trend. This study confirms that the choice of chamber used to assess honey bee drone sperm motility has a significant effect on the results wherein traditional slide coverslips may be contraindicated.
This work was supported by the Spanish MINECO (grant AGL2017-85030-R), and the DGA-FSE (grant A07_17R).

[P.16.409] Weight queen Africanized honeybees produced with single and double larvae grafting technique with protein supplementation

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The Africanized queen honeybee weight is a factor that can be used to establish colony productivity standards. In this way, we aimed to evaluate weight bees emergency queens produced with unfermented and fermented supplementation, associated technique single and double grafting. Thirty colonies of Africanized honeybees were divided into six treatments: 1) No supplementation with single larvae grafting; 2) No supplementation with double larvae grafting; 3) unfermented supplementation and single larvae grafting; 4) unfermented supplementation and double larvae grafting; 5) fermented supplementation and simple larvae grafting; 6) Fermented supplementation and double larvae grafting. The unfermented protein supplement was composed of sugar, isolated soy protein, beer yeast, honey, bee pollen, palm oil, flaxseed oil, soy lecithin, and vitamin nucleus.The fermented supplement had the same formulation, but after mixing ingredients, microorganisms of the genera Streptococcus, Lactobacillus, Saccharomyces, and Bifidobacterium were added in an anaerobic environment under controlled temperature conditions for multiplication and fermentation.Supplements were supplied to the colonies at the time of the larvae grafting in hive surface feeders. The newly emerged Africanized queen honeybees were desensitized with CO_2 recorded weights using 0.0001gprecision scale. The data were analyzed in a double factorial scheme and the means were compared by the Tukey's test with 5% probability. Queen honeybees had weighted (mg) of 161.50d; 181.52b; 173.90c; 198.01a; 175.04bc; 178.41bc, respectively according to the treatments above mentioned.The weight of queen honeybees presented a difference ($p < 0.05$), the heaviest (198.01 mg) produced with unfermented supplementation and associated with the double grafting technique. Queens lighter were observed in the treatment without supplementation with simple grafting. Thus Africanized honeybee colonies when using double larvae grafting method supplemented with protein sources produced heavier queens.

APITHERAPY

09 SEPTEMBER 2019

POSTER SESSION 05 08:30-18:00
APITHERAPY I POSTER AREA

[P.05.102] Evaluation of antibacterial Activity of the protein fraction of *Melipone beecheii* and *Apis mellifera* honeys against Foodborne Pathogens

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High incidence of acute diarrhea caused by microorganisms related to foodborne diseases has sparked the search for solutions to avoid this problem. According to the Word Heath Organization (WHO), there are 1.7 billion cases of childhood diarrhea each year, 525 000 of whom will end in the death of the affected person. An alternative to solve this health problem is the use of regional honeys from *Melipona beecheii* and *Apis mellifera*, which have been determined to have an antibacterial potential against various pathogens related to Foodborne Illness. Previous studies have shown that honey proteins participate in an important way to the antimicrobial activity of the same. Therefore, the objective of this work was to evaluate the antimicrobial activity of different protein fractions present in the honey from *Apis mellifera* and *Melipona beecheii* against Escherichia coli, E. coli O157: H7 and Staphylococcus aureus.
Honey samples were collected from hives in the state of Yucatan, Mexico. Strains used were E. coli ATCC 25922, E. coli O157: H7S and S. aureus 25923. The extraction of proteins from honey was carried out by ultrafiltration. The separation of the protein extract was carried out by affinity chromatography (concanavalin A). The proteome of honey proteins was determined using SDS-PAGE and 2D system. Moreover, the proteolytic activity was determined using electrophoretic methods (zymogram). The antimicrobial effect was evaluated by disk diffusion method and the minimum inhibitory concentration (microdilution method).
The protein extract of *Apis mellifera* and *Melipona beecheii* honeys showed antimicrobial activity against the three pathogens. Melipone honey proteins showed the highest antimicrobial potential, requiring concentrations lower than 4.2 µg/mL. Proteome analysis of Melipone and Apis honey by 2D electrophoresis showed a total of 24 and 20 different proteins respectively. Antimicrobial analysis revealed that the non-glycoprotein fraction can inhibit the growth of bacteria tested, furthermore it also showed contain a protease by substrate-gel electrophoresis.
Honey proteins contribute and have a direct correlation with the honey antibacterial activity. These proteins have antimicrobial activity against Gram positive and Gram negative bacteria. Non-glycosylated proteins fraction contain at least one protease that might be the main responsible compound for antimicrobial potential.

[P.05.103] Oxymel - medicine with honey and vinegar

G. Nedoma
Aesculus Og, Wolkersdorf Im Weinviertel, AUSTRIA

Oxymel is a medicinal honey and vinegar syrup. Anchored for 2,500 years as a separate dosage form, Oxymel is one of the most important and interesting historical apitherapeutics. The name Oxymel stands for the Greek terms sour (oxy) and honey (meli) and means sour honey or sour syrup. Several thousand pharmacologically active oxymel types have been documented in history. In the industrial age Oxymel slowly disappeared from Pharmacopoeias and today the original Apitherapeutikum is largely unknown. For millennial treated doctors and naturopaths such as Hippocrates of Kos (460-370 BC), Galen of Pergamon (129-216) and Hildegard von Bingen (1098-1179) countless diseases with Oxymel. The simple mixture of honey and vinegar is pharmacologically active and is called Oxymel simplex (simple oxymel). Also documented are numerous phytotherapeutics that included one or more plants. Oxymel gives many answers to health issues of our time. For children and adults, Oxymel provides access to a natural disease-focused natural medicine through an easy-to-use and safe-to-use compound. Possible applications for Oxymel:
Naturopathy Numerous traditional therapeutic indications for Oxymel have been documented, including respiratory diseases such as rhinitis, sinusitis, tussis, bronchitis, asthma, allergic reactions, pyrexia, indigestion such as gastroenteritis, diarrhea, reflux, intoxication, colic, Lebergallen disease, acidosis, vomiting, constipation, dyspepsia , Renal insufficiency, urinary retention, psychosomatic disorders such as depression, anxiety disorders and insomnia or skin wounds.
Pharmacon Oxymel is an excellent extractant and preservative and offers alternatives to non-alcoholic phyto-extracts, pediatric medicines, cough syrups, bitters and wound care products.
Food Supplements Oxymel is particularly effective as an isotonic, where it can be used to increase athletic performance, geriatrics, dehydration after vomiting, diarrhea and fever, as well as hot flashes. In the field of fasting and detoxification treatments Oxymel is a sensible measure for the removal of pollutants and metabolic products from the organism.

[P.05.104] Effects of honeybee venom on acne

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Acne vulgaris is a chronic dermatologic disease with four factors involved in the development of lesions. Treatments need to address as many of these underlying factors as possible in order to reduce acne lesions. As such Purified Bee Venom (PBVTM) serum is an attractive therapeutic option for acne, but little data exists on the efficacy of this treatment strategy. In this prospective, non-comparative study, 30 subjects having mild to moderate acne vulgaris were enrolled and treated with PBVTM serum twice daily for a period of 6 weeks. Clinical evaluation of lesions by expert visual grading and image analysis were made at weeks 0 (baseline), 3 and 6. The average visual acne grade of all volunteers significantly improved with the PBVTM serum treatment at weeks 3 ($p<0.05$) and 6 ($p<0.001$) when compared with the baseline grade at week 0. In addition, there was a mean percent improvement of 8.6% and 52.3% in acne grade observed after 3 and 6 weeks of PBVTM serum use, with 20% and 77% of the subjects showing improvement, respectively when compared with baseline. Moreover, the subjects showed improvement in open comedones, closed comedones, papules, pustules and nodules after 3 and 6 weeks of PBVTM serum use. Six weeks of treatment with PBVTM serum was found to be effective in the treatment of mild to moderate acne vulgaris, with no incidence of serious side effects or irritation.

[P.05.105] Bee venom crème - not everybody is a hero - The variety of applications for bee venom in form of creme

T. Gloger
Api-Zentrum Ruhr, Castrop-Rauxel, GERMANY

Bee venom is a wonderful therapeutic. But most people who could profit from BVT are refraining as they are frightened. A low barrier entrance with high convenience to BVT is the application in form of crème.
The classic applications had always been the pain relieving and anti-inflammatory action. But when leaving the paradigm that the bee venom must always penetrate deep into the body the number of applications widens. Bee venom with its antibacterial and antiviral properties is able to act on sensitive tissue, too. This way is similar to coming from bee stings to micro-stings. The formulation of the crème is key, but its application is much easier than micro-stings.
We found nearly 30 applications, including of course, cosmetic formulations targeting the antiwrinkle "bee tox instead of Botox".
Starting with mildish formulated cremes we can treat skin problems like acne due to their antibacterial and regenerative properties. For a neurodermitis or psoriasis skin cremes can work as anti-itching agents.
Formulations which are based on more oily components are useful for massage purposes e.g. foot reflex massage.
In the latest stage we are going to a more aggressive formulated version. This contains plenty of skin openers and is useful for the classic applications like pain, pain reliving. Here the anti-inflammatory property of bee venom is the targeted mode of action.
In a nutshell increasing the mode of action gives step by step different applications. This is important for apitherapy and might convince possible users in a gently manner that classical BVT could be useful for them. too.

[P.05.106] Use of 2% propolis dressings in complicated wounds of diabetic patients

L. Ghersi, N. Chirino
Centro Médico Cubano Habana Medic, Lima, PERU

Summary: Propolis are a set of resins that collect bees for antiseptic and protective of the hive. In the pharmaceutical industry has begun the use of propolis as a immunomodulatory agent, tissue regenerator and as a broad spectrum antibiotic. We selected a group of patients treated by the service of vascular surgery and angiology presenting diffuse phlegmon in lower limbs that have not been previously cured and/or have had complications in their injuries due to inadequate previous cures In the period 2015 – 2016.
Materials and methods: We used propolis impregnated gauze at 2% concentration In an oily base for the treatment of wounds in diabetic patients. 5 representative cases will be presented where the evolution of the lesions (tissue necrosis state, resurfacing, Neo vascularization and scarring) will be observed. The patients underwent prior intravenous antibiotic treatment. The healing of the lesions and change of the dressings was performed every 48 hours. The maximum treatment time during the study was 90 days.
Conclusion: It is concluded by the review of cases that propolis have a regenerative, antiseptic and healing effect on complicated wounds of diabetic patients.

[P.05.108] Immune potentiator of bee propolis

A. Hegazi [1], F. Al Guthami [2], A. Al Gethami [2]
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Propolis had been well documented in traditional medicine for treating systemic immune diseases, allergic diseases, viral diseases and organic-specific inflammatory diseases since more than one thousand years. During the last ten years, immunoregulatory and anti-inflammatory properties of propolis have been published. The therapeutic characteristics of propolis have been well known for a very long time. It has been used in folk medicine for different nations as early in Egypt as 3000 BC.

In recent years, propolis has become a subject of increasing interest for chemists and biologists. It had various biological and therapeutic activities. In in vitro and in vivo assays provided new information concerning its mechanisms of action. Propolis possesses variable biological activities activity was investigated on antiviral, antibacterial, fungicidal, anti-inflammatory, antitumor, antiparasitic, antioxidant, cytokines and immune response. Immunomodulatory and antitumor properties, considering propolis effects on antibody production and on different cells of the immune system which involving the innate and adaptive immune response. This review was aimed to through more light on the enhancing activity of propolis on immune response, and opens a new perspective on the investigation of propolis biological properties on immune mechanism.

[P.05.109] Dermatological care and beauty treatment (cosmetic) with bee products - honey, beeswax, queen cells wax, Propolis, N-Chromosome Royal Jelly, Royal Jelly and bee venom

N. Valiaftari, M. Chinaveh, M. Chinaveh
Caspian Apiaries, Delta, CANADA

With aging, tissue repair and cell regeneration slow down. The amount of natural moisture present in the skin is reduced. Because collagen production is less, the skin becomes thinner and loses its flexibility. In addition, malnutrition and exposure to extreme cold, sun, smoke, smog, and chemicals, as well as dehydration and sleep deprivation can damage skin and hair. Imbalanced hormone, such as low estrogen, makes the skin drier, increases wrinkles, reduces collagen and elasticity, and causes the thinning hair.

In order to have a well-nourished skin, we require to supplement our body with a sufficient and balanced amount of vitamins, minerals, proteins and carbohydrates.

Producing creams that contain absorbable proteins and minerals is difficult. Also, most ingredients used in beauty products require preservatives or chemicals.

Using a combination of the following bee derived products as an alternative, can provide highly absorbable nutrients. Also, as long as bee venom and/or Propolis is used, there would be no requirement for preservatives or chemicals.

Bee pollen, Royal Jelly and NCRJ are water soluble and their nutrients can easily be absorbed internally and externally. This combination contains the majority of vitamins, minerals, amino acids and carbohydrates that a person needs to maintain the health of the skin.

Queen cells wax, Propolis, bee venom and NCRJ are strong antimicrobial and anti-pathogenic. This combination, helps protect the skin from environmental damage, such as exposure to extreme cold, sun burn, smoke, smog, chemicals, and certain microbial pathogens.

[P.05.110] Improvement of Chemotherapeutic Agents by Algerian Propolis in Lung Cancer

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The purpose of our research is to elucidate the behavior of oxidative stress in the cancerous pulmonary cell during the combined treatment of Propolis and antineoplasic agents and to explore the efficacy of Algerian propolis in reversing multidrug resistance and sensitizing chemo-resistant lung cancer cells (A549/DOX) to chemotherapy with DOX. Therefore, the cancer lung has been induced by a single intraperitenally dose of benzo(a)pyrene 200 mg/kg. After 16 weeks, the animals have been treated with the intraperitenally dose of cyclophosphamide 600 mg/m2 + Epirubicin 80 mg/m2 + 5 Fluorouracil 600 mg/m2 after a pre-treatment by Propolis 25 mg/kg given orally for 5 days. The animals were sacrified after 52 days. Then, the oxidative status of pulmonary tissues, of the control and experimental groups were evaluated. To investigate the mechanisms of MDR reversal agents, intracellular accumulation of DOX and Pgp-pump activity were determineted.

Benzopyrene has been able to generate an oxidative stress that could be at least partially involved in the induction of the pulmonary cancer where the cancerous cells showed enzymatic and non-enzymatic antioxidants decrease and an increase in lipid peroxidation. Histological analysis of lung tissue further supported our findings and demonstrated that treatment with the antineoplasic agents in association with propolis, changes not only the antineoplasic activity, but also ameliorates this activity by reducing the side effects. Also, the Propolis inhibited in a concentration-dependent manner, the Pgp efflux-pump, enhancing thereby the intracellular level of DOX. These results seem to be particularly important for both their clinical applications and the possibility for their use in the potential chemotherapeutic.

[P.05.111] The promising antibacterial activity of some Saudi Arabia honeys

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[1] *National Research Centre, Giza, EGYPT,* [2] *Al Guthami Foundation, Jeddah, SAUDI ARABIA*

Aim: The aim of this investigation was to through more light on the promising antibacterial activity of some Saudi Arabia honeys.

Materials and Methods: Sex Saudi Arabia honeys were used against some antibiotic-resistant pathogenic bacterial strains. The bacterial strains were Staphylococcus aureus, Streptococcus pyogenes, Klebsiella pneumoniae, Escherichia coli and Pseudomonas aeruginosa.

Results: The potential antibacterial activity of Saudi honey against five bacterial strains showed different levels of growth inhibition according to the type of honey. The overall results showed that the potential activity was differing according the pathogen and honey type.

Conclusion: It could be concluded that the Saudi honeys inhibit the growth of bacterial strains and that honey can be used as complementary antimicrobial agent against selected pathogenic bacteria.

[P.05.112] The use of green Propolis for psoriasis rheum – a case study

T. Gloger
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Autoimmune-diseases are difficult to treat. In the case of rheum cortisone is used besides various medication e.g. like MTX (Methotrexate). The progression of the pain can be slowed down. But the inflammatory process cannot be stopped sustainably. Side effects are heavy as e.g. severe liver damages and other damages occur. In our case the patent with a psoriasis rheum collapsed despite close-meshed surveillance of not only liver enzyme values. He decided to treat further on with naturopathy. Step1 of this naturopathy was gut renovation with different bacteria e.g. effective microorganism (EM) and dietary fibres. In Step 2 food allergens were searched and carefully avoided by special diet. Due to improvement of the colon colonisation this is a fluctuant process and over time some of the food sensitivity relieved and the diet could be modified dynamically. Parallel to step 1 and 2 the patent started to take high dose propolis. Propolis fights against different kinds of inflammation and pain processes. In this case green, organic propolis from Brazil (from the state of Minas Gerais) was used.
The total process of recovery lasted 24 months. After that time the patient was free of pain. Only the already deformed joints could not recover any more. The affected finger and toe stand still out not naturally. Despite the malposition these joints evoke no pain.
Today the patent complies still strict to his diet and takes a maintenance dose of green propolis.

[P.05.113] Study of the topical solution of própolis as agent carrapaticida

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The species Canis familiaris is attacked commonly by parasites that grow in the derme, originating from of the infected environment or for the conviviality with other infested animals. The presence of the tick Rhipicephalus sanguineus, it can take the dogs develop her the disease of the tick- Erlichiose, being vectorial of the bacteria hemoparasitas rickétsias and Erlichia canis. The present work search to evaluate the therapeutic action of the própolis extract. propolina - in the extinction of the parasite. The própolis is a natural product of the bees, produced starting from vegetable resins. In the beehives the própolis is used by the bees to inhibit the development of acarids, mushrooms and bacteria, as well as to ban the box's openings and to minimize the wind entrance and rain. Many studies tell the action of the própolis as antibiotic and bactericidal agent in the treatment of throat infections, cutaneous wounds and in the treatment dermatológico of acnes. After the daily treatment in 12 dogs of you vary races, separate in 12 stalls, for 15 days, several ticks died or they fall of the host, being exterminated in adult number the youngest ticks, in I work as a trainee him of nymph. It is still done necessary a study of several propolina concentrations for better efficiency of the product in adult ticks.

[P.05.114] Amino acid substitution of mastoparan-B on antimicrobial potency against multidrug-resistant Escherichia coli

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Mastoparan-B (MP-B) is an antimicrobial cationic tetradecapeptide (LKLKSIVSWAKK VL-CONH2) isolated from the hornet venom of Vespa basalis. We found that amino acid-substituted MP-B analogs (MP-B-1 to 6) exhibited diverse antimicrobial activity against clinical isolates of multidrug-resistant Escherichia coli (MDR E. coli) and had variable physico-chemical properties. All six MP-Bs showed very low hemolytic activity on sheep erythrocytes even using as high as 256 mg/L of MP-Bs, suggesting that all with negligible cytotoxicity. Among MP-Bs, MP-B-1 ([Trp3]-MP-B) has better antimicrobial activity against E. coli isolates. Among these bacterial isolates, both MDR E. coli PFH12 and PFH14 were respectively detected containing 8 and 6 antimicrobial resistance genes, and indeed resistant to ampicillin, gentamycin and tetracycline. It has been demonstrated that antibacterial proteins show antibacterial activity against pathogens by changing cell membrane permeability. In this study, we also verified that both MP-B and MP-B-1 can cause membrane permeabilization on MDR E. coli isolates in a dose-dependent manner with low effective concentration 50 (EC50) values. Antimicrobial combination study showed that MP-B or MP-B-1 acted synergistically with ampicillin, gentamicin or tetracycline against MDR E. coli isolates. Some antimicrobial combination showing synergy, partial synergy or additivity also displayed rapid bacterial-killing activity against MDR isolates. It also revealed that at least 2-fold MIC decreased in MIC with both antimicrobial agents in these antimicrobial combinations. In conclusion, our study resulted that MP-B with an appropriate amino acid substitution could enhance antimicrobial activity and when combing with antibiotic also exhibit synergistic activity against E. coli. Accordingly, modification by amino acid substitution could be the feasible strategy for rational design of antimicrobial peptides to enhance antimicrobial activity.

[P.05.115] Anti-aging study of noble Bumblebee (Bombus terrestris) queen glycosaminoglycan in aged rat

M.Y. Ahn [1], H.J. Yun [1], J.S. Hwang [1], K.K. Park [2]
[1] *National Academy of Agricultural Science, Wanju-gun, SOUTH KOREA,* [2] *Pharmacogenechips Inc, Chuncheon, SOUTH KOREA*

The objective of this study was to evaluate anti-aging effect of a newly prepared glycosaminoglycan (GAG) derived from *Bumblebee* (*Bombus terrestris*) queen (BTQG, 10 mg/kg). Gryllus bimaculatus (Gb, cricket) GAG (GbG, 10 mg/kg) or glucosamine sulfate (GS) was used as a positive control. N-glycans derived from BTQG were composed of hexose polymer including Hex8, Hex3HexNAc3dHex2 and Hex5HexNAc3dHex2 as active ingredients. They were intraperitoneally administered to 14 month-aged rats for one month. BTQG reduced serum levels of free fatty acid, ALP, sGPT, creatinine, and BUN, showing hepato-and renal protective effects with anti-lipidemic activities comparable to GbG. Microarray results showed that BTQG, GbG or GS treated rat group commonly showed simultaneously upregulation of 36 genes compared to the control group, including secretogranin II, AP-1 regulated protein related ROS DNA damage repair, metallothionein 1a, and alpha-2 macroglobulin. They also showed 417 downregulated genes, including vimentin, moesin, and mitochondrial carbonic anhydrase. These genes might play a role in their anti-aging action.

[P.05.116] Elongation of survival of Bumblebee extracts on organ tissue cells in aged rats

M.Y. Ahn, H.J. Yoon, J.S. Hwang
National Academy of Agricultural Science, Wanju-gun, SOUTH KOREA

This study aims to investigate the anti-aging effect of a newly prepared *Bumblebee* extracts, given to aged rat organ cells for 2days incubation. *Bumblebee* extracts was found to be related to have anticancer and anti-fibrinolytic activities in *Bombus ignitus* larva extracts. We would like to consider that aging is phenomenon in which normal body cells can no longer divide after a certain number of breaks, contributing to aging of individuals and tissues; there by an important mechanism that inhibits abnormal growth in body's organs. While studying the various physiological activities of insects in the *Bumblebee*s, extracts of the *Bumblebee*s (B. terrestris, B. ignitus, B. hypocrita sapporoensis), and B. ardens, these Bumbus sp. was investigated to elongate survival and suppress inflammation factor IL-10 in 18 month-aged rat primary organ tissue cells from kidney, spleen and pancreas. Therefore, Bombus sp. extract involves with an aged body long-term protection agents characterized by the extraction of *Bumblebee* preparation.

APITHERAPY

11 SEPTEMBER 2019

POSTER SESSION 17 08:30-18:00

APITHERAPY II POSTER AREA

[P.17.411] Using N-Chromosome Royal Jelly (NCRJ) in combination with other bee by-products for diabetes and wound management; Based on studies performed on 889 patients in Iran, Canada and USA

H. Yeganehrad, S. Jazini-Dorche, S. Yeganehrad
Caspian Apiaries, Delta, CANADA

Diabetes is a chronic disease that can cause ulcer, infections that may lead to foot or leg amputation, blindness, kidney failure, heart attacks, strokes and heart failure. Today, 11 million Canadians live with diabetes or prediabetes.
This research was done in Iran, on 837 people, and in Canada and USA, on 52 people, with the majority of them requiring amputations, for the period of 8 years. Patients with type-2 diabetes were divided into 3 groups; all were using Metformin, Insulin and Glibenclamide.
Group A: Patients with severe infections, requiring amputation.
NCRJ in combination with honey, bee pollen and Propolis was given orally, 4 to 5 times, and bee venom, royal jelly and Propolis was used topically, 2 times, if required.
10 years ago maggots were used topically for infections and wounds, in combination with NCRJ (consumed). But due to patients' negative reactions and inefficiency, maggots were replaced by bee-by products.
Group B: Patients with bed ulcer, kidney failure and those requiring eye surgeries.
NCRJ, Propolis, honey and bee pollen were given to consume 4 to 5 times per day. Topical cream was used for bed ulcer or dry skin, 2 times per day.
Group C: Patients with early stages of diabetes.
NCRJ and Propolis were given 2 times per day.
The results were significant; 483 diabetic patients from Iran and 36 from Canada and USA, did not require diabetes medication after 3 months of treatment. The remainder reduced their diabetes medication over 80%. Blood sugar level, cholesterol and hormones (progesterone, testosterone and estrogen E1/E2/E3) became balanced, eye vision was improved – in some cases no further laser treatment was required, eczema, skin dryness, asthma and mineral deficiencies were treated.
In this presentation the significance of using NCRJ, honey, bee pollen, Propolis, and bee venom as a treatment for diabetes will be discussed in further details. After seeing the positive effects
That various research group and professional in the medical field to work together
Based on our observations, we see the importance of further clinical studies in order to affirm the positive effects of bee by-products on diabetes and autoimmune diseases.

[P.17.412] Effects of N-chromosome royal jelly supplementation on white blood cell count in children with acute lymphoblastic leukemia on chemotherapy

G. Bahoush Mehdiabadi, R. Pahlevan, H. Yeganehrad
Aliasghar Children Hospital, Tehran, IRAN

Background and objective: To our knowledge, no previous studies have focused on the increasing effect of fresh n-chromosome royal jelly (RJ) administration on white blood cell (WBC) count. Our aim was to study the effect of fresh n-chromosome RJ administration on WBC count in children with acute lymphoblastic leukemia (ALL) under the maintenance chemotherapy.
Methods: This was an open-label pilot study in which 20 children with ALL received 2g of freshly prepared n-chromosome RJ daily. The dose was increased every two weeks up to three times a day, if WBC count could not significantly increased.
Results: The mean age of all enrolled patients (11 boys and 9 girls) was 6.10 ± 2.3 years (2.1-12.4) at the time of the study. The mean WBC count before and after fresh n-chromosome royal jelly (RJ) administration were 2857.00 ± 411.49 And 4480.00 ± 579.23, respectively, (P = 0.035). In one patient, the count of WBC did not change and in another patient, the count of it slightly decreased. Some of the patients needed to increase dosage of n-chromosome RJ for increasing WBC count. There were not any significant side effects after this treatment.
Conclusion: This is the first human study on the effect of n-chromosome RJ supplementation in children with ALL. Our results showed significant increased WBC count with the treatment with regard to the clinical severity score and laboratory markers for the disease. At this stage, it is a single study with a small number of patients, and a great deal of additional wide-scale randomized controlled studies are needed to critically validate the efficacy of n-chromosome RJ in increasing WBC count among children with ALL.

[P.17.413] Effect of consuming of N-Chromosome Royal Jelly (NCRJ) on increasing White Blood Cell (WBC); based on research study performed at children's hospital in Iran, on patients under chemotherapy treatments

Z. Azarae, P. Pish Bahar, P. Azarae, H. Yeganehrad
Caspian Apiaries, New Westminster, CANADA

Certain chemotherapy drugs kill many of the cells in the bone marrow (bone marrow is responsible for making blood cells), which can put patients at the risk of low blood cell count (WBC, RBC, HB and PLT), which may lead to serious complications, such as delaying the treatments, liver damage, anemia and/or bleeding, and increased susceptibility to infections, which in severe cases may lead to death.
This research was performed in Aliasghar children's hospital in Iran, during 2017-2019, on children with various cancers, such as ALL (Acute Lymphoblastic Leukemia) and AML (Acute Myeloid Leukemia), from the age of 3 to 15, and with one case study on an 8-month infant, in 2016.
NCRJ was first used to increase blood cell counts of Parsa Pish Bahar, an 8-month infant, who was diagnosed with testicular cancer. Doctors removed the germ cell tumor and started the chemotherapy right away. After 6 periods of chemotherapy, the result of ABR test showed that Parsa was unable to hear sounds in the frequency of 20 Hz. - this was the side effect of Cisplatin drug.
Before the treatment, Parsa's WBC was normal, 8000 cells/mm3, on the 3rd day, reduced to 4000 cells/mm3 and on the 5th day reduced to 2000 cells/mm3. Between each cycle of treatment, Parsa spent 10 days at home, during which, his WBC increased to 7000-8000 cells/mm3 by consuming NCRJ.
After seeing this positive effect, the research began at Aliasghar children's hospital, in order to come to a definite conclusion. 30 children in 2017 and 61 in 2018/19, were chosen, who were under Cytarabine (Ara-C), Methotrexate and Vincristine, and were experiencing various side effects, such as low blood cell count and mouth sores. For each patient, 3 grams of NCRJ was given per day for one month. The WBC either stayed the same or increased; some children no longer required artificial WBC.
Results of laboratory tests for Amin, a 3-year old boy:
19 August 2018 (During Chemotherapy):
WBC: 700
RBC: 2500
HB: 7.4
Platelet: 55000
25 August 2018 (6 days after consuming NCRJ)
WBC: 4600
RBC: 3180
HB: 9
Platelet 131000

[P.17.414] Effect of consuming of N-Chromosome Royal Jelly (NCRJ) on increasing Platelets (PLT) of children affected by cancer; based on research study performed in children's hospital in Iran

P. Azarae, Z. Azarae, P. Pish Bahar, H. Yeganehrad
Caspian Apiaries, New Westminster, CANADA

Certain chemotherapy drugs kill many of the cells in the bone marrow (bone marrow is responsible for making blood cells), which can put patients at the risk of low blood cell count (WBC, RBC, HB and PLT), which may lead to serious complications, such as delaying the treatments, liver damage, and increased susceptibility to infections, which in severe cases may lead to death.
A low platelet count can lead to serious blood loss, which can damage internal organs.
This research was performed in Aliasghar children's hospital in Iran on children suffering from various cancers, such as ALL (Acute Lymphoblastic Leukemia) and AML (Acute MyeloidLeukemia), and under chemotherapy , from age 3 to 15. These children were experiencing side effects from chemotherapy drugs - Cytarabine (Ara-C), Methotrexate and Vincristine - such as low blood cell count (WBC, RBC, HB and PLT) and mouth sores.
NCRJ was first given to Parsa Pish Bahar, an 8-month infant, who was diagnosed with testicular cancer, in 2016. Doctors removed the germ cell tumor and started chemotherapy right away. Parsa experienced various side effects from Cisplatin drug, including low blood cell count (WBC, RBC, HB and PLT) and temporary damage to his ears – he was unable to hear sounds in the frequency of 20 Hz.
He was given NCRJ during his chemotherapy to treat these side effects; his blood cell count (WBC, RBC, HB and PLT) increased and his immune system was improved. After seeing this positive result, the research study began at Aliasghar children's hospital, in order to come to a definite conclusion.
3 grams of NCRJ was given to each patient, per day for one month. After consuming NCRJ, the patients did not require to have RBC and PLT transfusion and/or artificial WBC.
Amin, a 3-year old boy, is one the examples, below is the result of his laboratory tests:
19 August 2018 (during chemotherapy)
WBC: 700 cells/mm3

RBC: 2500
HB: 7.4 g/dL
Platelets: 55000
25 August 2018 (6 days after consuming NCRJ)
WBC: 4600 cells/mm3
RBC: 3180
HB: 9 g/dL
Platelets: 131000

[P.17.415] Effect of consuming of N-Chromosome Royal Jelly (NCRJ) on increasing Red Blood Cells (RBC); based on research study performed at children's hospital in Iran, on patients under chemotherapy treatments

P. Pish Bahar, Z. Azarae, P. Azarae, H. Yeganehrad
Caspian Apiaries, New Westminster, CANADA

Certain chemotherapy drugs kill many of the cells in the bone marrow (bone marrow is responsible for making blood cells), which can put patients at the risk of low blood cell count (WBC, RBC, HB and PLT), which may lead to serious complications, such as delaying the treatments, liver damage, anemia and/or bleeding, and increased susceptibility to infections, which in severe cases may lead to death.
This research study was performed in Aliasghar children's hospital in Iran, during 2017-2019, on children suffering from various cancers and under chemotherapy treatments, such as ALL (Acute Lymphoblastic Leukemia) and AML (Acute MyeloidLeukemia), from age of 3 to 15, and with one case study on an 8-month infant, in 2016.
NCRJ was first used to increase blood cell counts of Parsa Pish Bahar, an 8-month infant, who was diagnosed with testicular cancer. Doctors removed the germ cell tumor and started the chemotherapy treatment right away. During the treatment, Parsa's immune system weakened and his RBC count and the level of Hemoglobin (HB) reduced.
After seeing the positive effect of NCRJ on increasing blood cell count, the research study began at Aliasghar children's hospital in 2017, in order to come to a definite conclusion. 30 children in 2017 and 61 in 2018/19, were chosen, who were under Cytarabine (Ara-C), Methotrexate and Vincristine, and experiencing various side effects from these drugs, such as low blood cell count and mouth sores. For each patient, 3 grams of NCRJ was given per day for one month. After consuming NCRJ, the patients did not require to have RBC and PLT transfusion and/or artificial WBC.
Amin, a 3-year old boy, is one the examples, below is the results of his laboratory tests:
19 August 2018 (during chemotherapy)
WBC: 700 cells/mm3
RBC: 2500
HB: 7.4 g/dL
Platelet: 55000
25 August 2018 (6 days after consuming NCRJ)
WBC: 4600 cells/mm3
RBC: 3180
HB: 9 g/dL
Platelet 131000
The results gathered from laboratory tests show that by consuming NCRJ, blood cell count increases and patients' immunity system will improve. However, further study is required to reach a firm conclusion.

[P.17.416] The importance of using bee products, N-Chromosome Royal Jelly (NCRJ), Bee Pollen, Propolis and Queen Cells Wax, for the treatment of patients with diabetes, suffering from various health conditions

M. Moradi Chamachar, G. Bahoush, H. Yeganehrad, R. Pahlevan
Aliasghar Children Hospital, Tehran, IRAN

Diabetes patients may suffer from various health conditions from high blood sugar, insufficient or lack of insulin production by Beta cells, protein deficiency, autoimmune diseases, which affect some of the internal organs such as liver and kidneys, and bed/foot ulcers and infections.
This study was done in Iran, Canada and USA, on 837 diabetes patients in need of amputation, according to the medical team. The treatment included both topical cream/liquid and oral intake, to treat ulcers/infections, as well as, the root causes of various health conditions the diabetes patients were affected by.
One of the major concerns in patients with diabetes is amputation which is caused by low blood circulation and infections. In some cases, the infection can even lead to death. Also, some of the pathogens and microbes are resistant towards antibiotics. Some antibiotics are ineffective due to poor blood circulation.
By supplementing patients with NCRJ (which was the main ingredient of the oral treatment), pollen, Propolis, and honey, blood circulation

was normalized, Beta cells started producing insulin, and the immune system was boosted, which helped the body to fight the infections. After this treatment, 80% of the patients did not require insulin, and with the exception of a few patients, the rest did not require amputation anymore.
In this method of treatment, not only the blood sugar became balanced, but also, the results of our tests, taken from before and after the treatment, showed that all other conditions including mineral deficiency, low blood cell counts, high blood pressure, fatty liver, and various infections, were improved significantly and some completely treated.
The above conclusions are based on observations and laboratory results, so, further clinical studies are required to come to a more stable conclusion.

[P.17.417] Combinations of Queen Cell Wax, Beeswax, Propolis with N-Chromosome Royal Jelly (NCRJ) to cure fee wounds and dry skin

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90%of adults that suffer from dry, cracked feet will have a microbial issue, such as the presence of a fungus, suffer from eczema or have excessively dry skin. Combinations of queen cell wax with regular wax, NCRJ and Propolis in a cream base have been found to help heal the open wounds, and provide a barrier against debris and bacteria while healing is occurring. Most studies that have been done on these products are to find out why this combination leads to success with various individuals. Three major factors were identified. The average human body temperature is 37 C, ad as beeswax can only melt at 60 C, it is able to create anaerobic conditions on the skin, which reduces the amount of aerobic bacteria or fungi found within the wounds. NCRJ, Propolis and queen cell wax are also known to contain compounds that can prevent infection. The combination of moisture with the queen cell wax creates antimicrobial compounds, further preventing reinfection. Finally, the presence of wax can act as a barrier from debris and microbes, allowing for new skin growth to occur. Beeswax used for similar purposes thousands of years ago in old Persia, not only on humans but on livestock and animals as well.

[P.17.418] Oral treatment of royal jelly improves memory and presents neuroprotective effects on an experimental model of Alzheimer´s disease

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Alzheimer's disease (AD) is a neurodegenerative disorder that is the most cause of dementia in the world. Currently there is no definitive treatment for AD and the available treatments only delay progression of the disease and control some symptoms. Royal jelly (RJ), a substance produced by worker honeybees of the *Apis mellifera* species, has been popularly used for more than 30 years in areas related to health eating and natural medicine. Researches indicate that RJ has a series of
pharmacological activities, including neuroprotective and improvement of cognitive function. The aim of this study is to investigate the effects of oral treatment of royal jelly in rats submitted to icv-STZ model of AD on learning and memory and neuroprotection, which is assessed by neurodegeneration, and oxidative stress. The icv-STZ model shows many aspects of SAD abnormalities, resulting in decreased brain glucose and energy metabolism, cognitive impairment, oxidative stress, astrogliosis, neuronal loss, and amyloid angiopathy. In this study, icv-STZ injection induced deleterious effects in the hippocampus, associated with cognitive impairments, and developed marked neurodegeneration, besides the reduction of neurogenesis and increased oxidative stress. On the other hand, RJ long-term oral administration induced beneficial effects in animals injured by icv-STZ injection, increasing retention time for working spatial memory, reducing neurodegeneration and oxidative stress level and increasing the proliferation of new neurons in the hippocampus. Thus, RJ promotes beneficial effects on cognitive functions and exhibits a neuroprotective action in the STZ experimental model of SAD.

[P.17.419] Apitherapy Statistics -2011-2017 from the Center for Alternative Therapies and Health Guidance (C.E.T.A.O.S) in Torreón, Coahuila, Mexico

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Apitherapy is the therapeutic use mainly of apitoxin. Endorsed by rigorous clinical studies, there are those who have suggested that it can treat a wide variety of ailments and diseases, especially those of autoimmune and degenerative nature, apitherapy is a type of complementary and alternative medicine that uses the derivatives of the hive, for its healing purposes. The objective of the present investigation was to evaluate and classify the different diseases treated through apitherapy in the Center for Alternative Therapies and Health Guidance (CETAOS) in Torreón, Coahuila, México. The analyzes were carried out with the Microsoft Excel program (Windows® Office 2013) for each of the years attended with apitherapies, averages were obtained and thus in percentages and represented graphically. The most common ailments found in the years 2011-2017 were the following: Localized pain, autoimmune system problems, nervous system, respiratory, circulation problems, spine and skin. The number of female and male patients per year was: in 2011, 60% were women and

40% men; in 2012, 75% women and 25% men; in 2013, 67% women and 33% men; in 2014, 69% women and 31% men; in 2015, 69% women and 31% men; 2016, 62% women and 38% men and, in 2017, 67% women 33% men. In this period, 748 (65.8%) women and 388 (34.2%) men were attended for a total of 1,136 people, so previously found the use of apitherapy to treat diseases or conditions is recommended to treat and cure various diseases.

[P.17.420] Evaluation of apitherapy as a biotechnics for the treatment of human diseases

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The apitherapy is a biotechnics - or ecotechnics - in which different beehive products are used for health benefits mainly the treatment with bee venom (apitoxin) is one of the most used, thanks to the results and benefits that this treatment bring for health. It is performed through apipuncture which stimulates specific parts of the body, through the application of bee stings in order to improve conditions suffered by people such as stress, sciatica, embolism, cysts, arthritis, goiter, muscle pain, varicose veins, paralysis, carpal tunnel, vitiligo, vertigo, multiple sclerosis and lupus erythematosus; In the present work, the response of people treated for these conditions in the Center for Alternative Therapies and Health Guidance (CETAOS) at the Universidad Autónoma Agraria Antonio Narro, Unidad Laguna, Torreón, Coahuila, México and the degree of satisfaction that people feel according to the application of this therapy. The registration of the people who attended and the pathologies to be treated was captured in the Excel statistical package (Windows® Office 2013). A questionnaire was applied to a population of 50 people by calculating the sample number (500 people in 6 months), which had six questions about how they felt with the treatment and with the help of the Likert scale, took performed a data analysis with the non-parametric rank test to compare the evolution of treatment considering before and after apitherapy. The comparison was carried out with the Wilcoxon.05 test with the Statistical Package for Social Science (SPSS® IBM) and it was found that there was improvement and satisfaction to the treated patients (n = 50, Point probability 0.043).

[P.17.421] Study of food safety on honeybee drone pupas

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In this study, safety investigations on harmful microorganisms and mycotoxins were conducted on honeybee drone pupae as a new food material, which are capable of being mass produced in apiaries and are rich in nutrients. The honeybee drone pupae produced in apiaries from three regions in Korea were collected and immediately frozen. Next, the samples were subjected to freeze-drying. According to the Korean Food Standards Codex test method, coliforms, Salmonella spp., Staphylococcus aureus, and enterohemorrhagic Escherichia coli were not detected in 280 honeybee drone pupas. In addition, in terms of mycotoxins, aflatoxin B1, ochratoxin A, deoxynivalenol, andzearalenone were not detected in honeybee drone pupas. Therefore, the honeybee drone pupae collected from the beehives and immediately stored frozen were safe from harmful microorganisms and mycotoxins. Thus, they can be used as food material.

[P.17.422] Study of Bee venom effect on Carrageenan induced paw inflammation model

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Objectives: The aim of this study was to investigate the efficacy of Mongolian honey bee venom (*Apis mellifera L.*) in carrageenan induced acute paw inflammation.
Methods and Materials: The study was carried out in Pharmacological center of the Institute of Traditional Medicine and Technology. Effect of bee venom on carrageenan induced paw inflammation was studied by determining paw edema, Plasma MDA, tissue MDA and cytokines such as TNF-alpha, IL-1beta, IL-6, IL-10. 40 experimental Wistar rats were divided into 4 groups: 1 intact group; 2 control group; 3 comparative group (indomethacin treated); 4 bee venom treated group (BVT).
Statistical results were analyzed by SPSS 20.0 and Graphadprism 5.
Result: Compared to the intact group, a paw inflammation in control group increased by 40.9% at minute 30, by 51.4% at minute 60, by 49% at minute 120, and by 68.5% at minute 240. Which shows that CR induces exudation dominated paw inflammation. Paw inflammation in BVT group 2.3 times less compared to control group at minute 240.
Compared to the control group, MDA levels in plasma of BVT group was lower by 15.5% and MDA levels in tissue was lower by 15.1%. BVT group's TNF-alpha level was lower by 9.5% compared to the control group. Also compared to control group, IL-1beta and IL-6 levels in BVT group was lower by 31.2% and 8.8% respectively. Hence, IL-10 in BVT group increased by 58.4%.
Conclusion:
1. Bee venom reduces the amount of swelling of paw edema in experimental animals.
2. Bee venom reduces the inflammation markers such as MDA, TNF-alpha, IL-1beta, IL-6.
3. Bee venom increases anti-inflammatory cytokine IL-10.
We can conclude that BV has anti-inflammatory effect in CR-induced paw inflammation

[P.17.423] Review on Medicinal Value of Honey in Ethiopia

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Ethiopia is endowed with variety of ecological and climatic condition. It is also home to some of the most diverse flora and fauna in Africa. The forests and woodlands of the country contain diverse plant species which is home to nectar and pollen. In Ethiopia, honey is the most widely used hive products serving many functions, ranging from local beverage called tej (honey wine) to different medicinal uses since ancient times. Ethiopians used honey for medicinal purposes as well, where bee venom was used to treat joint pain from arthritis and headache by exposing themselves to bee sting and mixing honey with other medicinal plants in traditional medicine

In recent days, however it was subjected to laboratory and clinical investigations by several researchers and it has found a great place in modern medicine. Honey has been reported to have an inhibitory effect on around 60 species of bacteria, some species of fungi and viruses. Honey and other bee products are used to treat illnesses and their symptoms as well as pain from acute and chronic injuries. Illnesses that honey may treat include diabetes, multiple sclerosis, arthritis, infections, shingles etc. And also can treat Injuries like wounds, pain, burns and tendonitis. In modern medicine treatment, honey may be applied by taking orally, applying directly on the injured body part and injected directly into the blood

[P.17.424] Bee collected pollen with enhanced health benefits, produced by fermentation with a kombucha consortium

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Background: The bioavailability for humans of the pollen bioactive compounds is limited.

Aims: Our aim was 19 to enhance the health-related benefits of the pollen by fermentation with a Kombucha / SCOBY 20 (symbiotic culture of bacteria and yeasts) consortium.

Material and methods: We performed the fermentation of pollen suspended from the beginning with SCOBY on sweetened green tea or on Kombucha vinegar, by adding pollen after 20 days of Kombucha fermentation. We analyzed: formation of bioactive compounds (anti-oxidant polyphenols, soluble silicon, hydroxy-acids, short chain fatty acids - SCFA); parameters related to Kombucha fermentation (dynamics of lactic acid bacteria - LAB, formation of organic acids, soluble sugar evolution on Kombucha vinegar); the influence of Kombucha fermentation on pollen morphology and ultrastructure; in vitro cytotoxic and antitumoral effects of the Kombucha fermented pollen.

Results: The pollen addition increases LAB proportion in the total number of SCOBY microbial strains. SEM images highlight the adhesion of the bacteria from SCOBY to pollen. Ultrastructural analysis reveals the release of the pollen content.

Conclusion: The content of bioactive compounds (anti-oxidant polyphenols, soluble silicon species and SCFA) is higher in the fermented pollen and the product shows a slight antitumoral effects on Caco-2 cells. The health benefits of pollen are enhanced by fermentation with a Kombucha consortium.

[P.17.425] Evaluation of EPP-AF® extract for the development of a face mask for acne treatment

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Acne is an inflammatory process that affects millions of young people around the world. One of the main causes of acne is the uncontrolled proliferation of Propionibacterium acnes bacteria. Currently, acne treatments are made with topical retinoids, topical antimicrobials, systemic antibiotics, hormone treatments for women, oral isotretinoin, benzoyl peroxide, topical azelaic acid, among others. Although effective, many of these treatments have considerable side effects. In the meantime, new alternatives of molecules that aim to reduce the causes of this disease are being studied in order to develop new acne treatments. Among these molecules, we can include propolis, a natural product produced by bees through mixing saliva and beeswax with exudate from different botanical sources. Several biological properties of this compound have already been demonstrated as its antimicrobial, anti-inflammatory, wound healing and antioxidant. Thus, we have evaluated the anti-inflammatory and antimicrobial activities of Brazilian Green Propolis EPP-AF® in the model of acne in order to develop an alternative to current acne treatments. Our results showed that EPP-AF® propolis extract was effective in preventing the growth of P. acnes, thus confirming its antimicrobial activity. In relation of its anti-inflammatory property, we verified that green propolis was able to induce an increase in IL-10 regulatory cytokine production while inducing a decrease in the proinflammatory cytokine IL- 6 at treated cells, thus demonstrating its immunomodulatory role in this model. The TNF-a levels was also measured, and although no statistical difference was observed, its presence at the inflammation site is positive, since it acts on the recruitment of immunity cells which may contribute to resolution of local inflammation. In addition, in some cases, the presence of TNF-a at the inflammation site also

promotes the later generation of the anti-inflammatory IL-4 cytokine by the local cells. Finally, the safety of EPP-AF® propolis extract was tested and no toxicity was found. So, our results demonstrate that this extract can be used in a promising new formulation for acne treatment and allow us to follow to clinic trials aiming to test a propolis face mask in patients with this disorder.

[P.17.426] Bactericidal effect of Chilean monofloral honeybee from Fagopyrum esculentum against multiresistant enterobacteria

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Antimicrobial resistance is the phenomenon by which a microorganism is no longer affected by an antimicrobial to which it was previously sensitive, allowing certain microorganisms (for example bacteria) to neutralize the effect of medications, such as antibiotics. The resistance is explained by both, bacterial mutation and acquired antibiotic resistance genes. At this point, honeybee, a tested natural bactericide is capable of eliminating sensitive and antibiotic resistant bacteria. Monofloral honeybee obtained from Fagopyrum esculentum (buckwheat) has shown high levels of polyphenols, increasing its antibiotic effect compared to a multifloral honeybee. The present study was carried out to determine the efficacy of Chilean monofloral honey from Fagopyrum esculentum on bacterial inhibiting. For that, bacteria such as Streptococcus pyogenes, Staphylococcus aureus, Escherichia coli, Pseudomonas aeuroginosa, Salmonella typhimurium multidrug-resistant (MDR) and Salmonella typhimurium strain 14028s, were cultured in Petri dishes including specific nutritional support in each case, including also four wells filled honeybee, one no diluted and the rest diluted 1/2, 1/4 and 1/8. The results showed higher average inhibition halos for Salmonella Typhimurium MDR and Salmonella Typhimurium strain 14028s (26.60 and 26.08 mm, respectively), compared to those from Pasteurella aeruginosa and Escherichia coli (14.00 and 13.66 mm, respectively). Staphylococcous aureus and Streptococcus pyogenes showed intermediate values (20.66 and 17.66 mm). It concluded monofloral honeybees from Fagopyrum esculentum influenced size of bacterial inhibition halos, showing a higher potential against Salmonella. Further studies including other monofloral honeybees and other bacteria should be done to validate the results.

[P.17.427] Effect of Royal jelly on the antioxidant enzymes activity following experimental varicocele in rat

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Varicocele is one of the most prevalent causes of infertility. Oxidative stress due to varicocele has an effect on the testis tissue and often leads to infertility in men. The present study aimed to determine the protective effect of royal jelly on antioxidant enzymes activity (catalase, SOD, GPx) and total antioxidant capacity(TAC), after induction of varicocele in rat. Twenty-one adult Wistar rats were divided into three groups. The control group (I), Varicocele and treatment with normal saline (II), varicocele and treatment with RJ (III). At the end of the experiment, all the animals were sacrificed and testes excised. The activity of catalase (CAT), superoxide dismutase (SOD), glutathione peroxidase (GPx) and TAC were measured. There was a significant ($p<0.05$) increase in the activity level of CAT, SOD, GPx and of the experimental varicocele and treated with royal jelly when compared activity of the experimental varicocele and treated with normal saline. Therefore, RJ is a potential area for further studies and improving in spermatogenesis cycle after Varicocele.

[P.17.428] Innovation in apitherapy: the tropical deseases in the center of diabetology with the natural plants of Bangoua

E.C. Nyonta
ETS John Carafa Retraite A Bangoua, Bangangte, CAMEROON

Intorduction: i am happy to receive a slice of antenna on this tray of apimondia that i know well and i am first of all thank his president. i am a retired nurse and a trained bee-keeper. it is by internet that i discovered apimondia that sold me the dvd of apitherapy in 2006. from that moment, i began to take care of many patients with the venom of bee by direct sting called again apipuncture. i am the 1st patient of the center.
The apipuncture on the malaria the venom of bee treats and warns this dangerous illness under tropics:
- for the prevention make 1 or 2 stings every month is sufficient.
- for the treatment a sitting of 5 to 10 stings is sufficient to clean the blood of all plasmodium. after this sitting the patient shivers and perspires a lot and at the end of this reaction the patient is healed.
According to the specificity of any organism, this bee venom can remain active several weeks or several months in the blood, and the patient during all this time doesn't present any more symptoms of the malaria. thus, a 49 years old woman of the bangoua village confided us in a report copied on a dvd support that she used to make his crisis of malaria every month; but since 2012 that she underwent 07 sittings of bee venom to our center, she hasn't suffer from malaria till today.
Thus, we can deduce that the bee venom destroyed the resistant stumps of plasmodium in the blood. it is also for this reason that we recommend the research of the vaccine of the malaria in the cool venom of bee; it will be very efficient.
Since the venom of bee warns and treat cancers and rheumatisms, it will therefore be a vaccine to large specter against all these illnesses.

[P.17.429] Chemical composition and in-vitro analysis of propolis samples collected from three vegetation zones in Nigeria on some pathogenic organisms

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The study examined the in-vitro antimicrobial activity of ethanol extracted propolis sample on some disease-causing organisms. The aim of the study was to determine the effect of different vegetation zones on the efficiency of ethanol extracted propolis on some pathogenic organisms. Both the physicochemical and phytochemical properties of the ethanol extracted. Propolis samples were also determined, using appropriate laboratory methods. The Agar Diffusion method was used for the antimicrobial screening of ethanol extracted propolis on some of the pathogenic organisms such as Bacteria (Staphylococcus cureus, Streptococcus mutans, Salmollena typhii and Pseudomonas aeroginosa) and Fungi (Candida albicans, Rhizopus, Aspergillus fumigatus and Fusarium peritonitis) obtained from pathology laboratory of National Root Crops Research Institute, Umudike. Results obtained showed that Propolis collected from Central Guinea Savannah had the highest amount of fat (61.68 ± 1.201) but the least amount of food protein (1.15 ± 0.02), while Derived Savannah Propolis contained the highest amount of crude protein (1.73 ± 0.032). Derived Savanna Propolis had the highest amount of Potassium (68.38 ±0.052). Rain Forest Propolis had the highest amount of tanning (1.63 ± 0.015). Flavonoid content varied significantly at $P < 0.05$ probability level amongst the vegetation zones, with a range of 0.48 to 0.61 mg/ml. Central Guinea Savanna zone had the highest amount of flavonoid (0.61 ± 0.15). Vitamin A content, varied significantly among propolis from different vegetation zones at $P < 0.05$ level of significance. Derived Savanna Honey contained the highest amount of Vitamin C (4.75 ± 0.040 mg/ml) and Vitamin B6 (0.60 ± 0.011 mg/ml). The result of the antimicrobial activity of propolis samples from the different vegetation zones showed a significant level of difference at $P< 0.05$ probability level. The Rain Forest propolis had the highest zone of inhibition on Pseudomonas aeruginosa and also on Aspergillus fumigatus (6.31 ± 0.076). On average, the chemical composition and the antimicrobial activity of Propolis on some pathogenic organisms are affected by differences in vegetation zones to varying degrees.

[P.17.430] First scientific studies of beehive air composition

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Within the last few years, “beehive air inhalation” has become more and more popular in apitherapy. Especially respiratory illnesses, such as bronchitis or asthma, can be improved by beehive air treatment. So far, clinical and scientific studies are almost completely absent. Therefore, beehive air therapy is not recognized as an alternative cure in Germany. Scientific investigations are still required.
In the bee season from May to August 2017, volatile organic components (VOCs) of beehive air were analyzed at the Dresden University of Technology using various “air sampling” techniques. For that purpose, the established GC-MS method for the analysis of honey-flavoring substances could be adapted to the new matrix “beehive air” [1]. The optimized air sampling technique’s manual solid phase micro-extraction (SPME) and thermal desorption tubes (TD) - proved to be suitable systems for extracting the VOCs from beehive air. Over fifty different VOCs were detected with the two systems (SPME & TD). The assignment of the VOCs to the individual bee products from the hive showed that the substances identified in the beehive air predominantly originated from propolis and beeswax. The lowest number of VOCs was detected in pollen, honey, drone brood, and royal jelly.
A first quantification of ten VOCs using TD-GC-MS revealed very low concentrations ranging from 0.08 to 4.57 ng/L beehive air. For some of the detected VOCs, air guide-line values from the Federal Environment Agency exist in Germany for indoor air. Exceeding the guide-line values may cause some individuals to react with headaches and irritation of the mucous membranes [2]. However, all the quantified VOCs in the beehive air inhaled directly by patients via a breathing mask during treatment were clearly below the specified indoor guideline values.

[P.17.431] The Use of Propolis-Pollen Ointment in Burn Wounds Treatment

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Apitherapy, the medical use of honey bee products, has recently become a rapidly growing branch of science. Bee products have many versatile therapeutic properties. One especially interesting bee product is propolis. It is a type of “bee glue” and may be effective in treating burn wounds.
The aim of this research, was an attempt to prove that a propolis-pollen ointment provided better treatment for burn wounds compared with Argosulfan treated wounds. The third group of wounds was not treated with any medicine or ointments. The research was conducted on two Polish White Lop-eared pigs. There were 27 contact burn wounds inflicted on the pigs, following the Hoekstra methods.
20 days after wounding the pigs, the wounds treated with propolis-pollen ointment were fully covered with pink epithelium and there was a significant reduction in the size of the wound surface. In the control group there were still dry crusts present. On day 23 it was noted that complete wound healing had taken place with the propolis-pollen ointment. The surfaces of the wounds in the sub-group treated with

Argosulfan were smaller in size with small, crusty pieces falling from the surface. The wounds in the group that received no treatment with any medicine were covered with dry, hard crusts and the wound size remained unchanged.
The final results concluded that the healing process of burn wounds treated with propolis- pollen ointment is much faster compared to the wounds dressed with either Argosulfan or with no treatment at all. The emerging scar in subjects appeared correct and there weren't any hypertrophic scars.
Our results proved that an apitherapeutic treatment such as propolis-pollen ointments was more effective in skin damage therapy compared with Argosulfan, the golden means in treating burn wounds. The treatment of a burn wound is a serious therapeutic problem and there is lack of published literature on the effectiveness of propolis-pollen ointments in the treatment of burn wounds. It would be wise to pursue further research in this direction.

[P.17.432] Antifungal Activity of Microcapsules Propolis from *Tetragonula spp* with Maltodextrin and Gum Arabic Through Spray Drying

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Propolis is a mixture of resin and saliva of Tetragonula spp. that have antifungal activity. The purpose of this study was to develop the formulation of microcapsules propolis as active ingredients with samples of rough (taken from outside the beehives) and soft (taken from inside the beehives) propolis type. Then test the antifungal activity of propolis microcapsules to Candida albicans. The research was carried out by making propolis microcapsules using the spray dry method using maltodextrin and gum arabic coating. The resulting microcapsules were evaluated and tested the antifungal activity against Candida albicans. The results of rough and soft microcapsule characterization showed that propolis microcapsules have powder form; typical of propolis, and has a brownish yellow color. The particle shape is uniform and round and has a particle size 9.32 μm - 14.61 μm; encapsulation efficiency of 11.22% - 83.04%; moisture content 5.58% - 11.84%; water solubility 98.19% and 98.31%. The in vitro test of the antifungal activity on Candida albicans with the highest inhibitory power diameter of 10 mm and 7.33 mm shows bee propolis microcapsules Tetragonula spp. has the potential as an antifungal. This study suggests that propolis has the potential candidate to develop as an antifungal drug.

[P.17.433] Apitherapy honeys from medicinal beekeeping

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Honey is a common, natural sweetener present worldwide. It is obtained from honeybees, which process the nectar of flowers or the secretions of living parts of plants. Honey has been historically used as traditional medicine for the treatment of various ailments and diseases. In the works by Ibn sina (Avicenna), Aristotle, Pliny, Dioscorides, Galen, Hippocrates, Varro and other ancient scholars there are traces and backgrounds on apitherapy, the therapy based on bee products.
Honey composition varies from one country to another. Iran is one of the good producer of honey in the world. Some parts in Iran are an ideal source of medicinal honey due to different climates, high mountains and medicinal herbs. Moreover, mountainous areas with dry climate rich in medicinal herbs provides ideal environment for producing honey of high quality. Its climate varies a lot with the existence of nearly four seasons.
We established a new bee farm in the middle east where developed the medicinal beekeeping by managing and characterizing our products by updated analysis methods which added more values to our products commercially also. In this project many criteria were focussed such as bee farm region, flora sources, instrument materials, hygienic treatments, storage room conditions, biological management and honeybee health. In this honey production, the high sugar complex concentration, hydrogen peroxide, the low pH, existence of phenolics and enzymes were determined and accelerated in the laboratory. Several chromatography techniques were used to identified the medicinal compounds which characterized our honeys. HPLCMS/MS, GCMS and LC-MSITTOF characterized the medicinal molecules inside of the honeys. Generally, main part of apitherapy is a medicinal beekeeping where the pure product would produce and can be characterized in the laboratory for overall uses. Honey, a complex natural tonic, can be used as organic food with antioxidant contents which improves and maintains human and animal health. The good antioxidant effect, sterility and easy availability of honey make it an ideal source of natural antioxidants.

[P.06.117] Work organisation in professional beekeeping: assessment method and consequences of management choices

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As in the case of most agricultural activities, beekeepers have to adapt their activity to a changing context while continuing to match their own expectations, especially in terms of income and work-life balance. The work organisation plays an important role in meeting this challenge, as it is both a component of the adaptive capacity of the farm and an important aspect of the beekeeper's quality of life. In the case of beekeeping, the work organisation is all the more important given that this is a highly seasonal activity: most of the work related to the hive management, to honey production or to pollination services is concentrated over a few months, resulting in a high workload during this period. Thus, a better understanding of this work organisation and especially of the possible consequences of some management practices or commercial choices on the beekeeper's workload will be useful to both current and future beekeepers.

To identify the main issues of work organisation on beekeeping operations as well as the possible consequences of some technical or commercial choices on this organisation, we adapted the Work Assessment Method (a method from INRA and "Institut de l'Elevage" research institutes), which was developed and is currently used by livestock researchers and advisors, to beekeeping. Through an interview with the beekeeper after the beekeeping season, this method provides an assessment of the workload per fortnight over the whole year and of the available time, and identifies the distribution of the workload among the workers of the farm.

Using this adapted method, fifty interviews with beekeepers were conducted in France in 2017 and 2018. That allowed us to identify patterns of work organisation of professional beekeepers and the consequences of some of their commercial or technical choices, such as queen breeding, on their workload and work organisation. Besides presenting these case studies, this presentation aims at outlining the assessment method and its possible uses in beekeeping development and as an analytical tool for advisors and beekeeping extension services.

[P.06.118] The Perplexity of Beekeepers & Honey-Bee Industry in West Java

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Here in West Java, Indonesia, honey making business is prominent because our climate advantage, but unfortunately the supply and demand stream isn't met yet, because: (1) Nowadays, the government does not dispose proper support to boost the local business of beekeeping despite demand of honey is high and agriculture sustainability issue related to bee's existence are rising. (2) Beekeeping still rather a traditional unpopular profession. As found in our survey, among 80 citizens 57,5% had certain prejudices against bees. Those range from insufficient profitability, to fear of bee stings, to a lack of knowledge on the importance of bees as pollinators.

Together with European Union, within Horizon 2020 program by European Commission in order to contribute in achieving UN Sustainable Development Goal (SDG N°2), "End hunger, achieve food security and improved nutrition and promote sustainable agriculture", we are doing research on beekeepers behaviours in Indonesia. This research is to contribute in developing SAMS (Smart Apiculture Management Service) that aim to give benefit for existing beekeepers in bee health monitoring, managing beekeeping activities, and eventually boosting Indonesian apiculture to be sustainable. In order to develop a user-based, effective, and sustainable system of SAMS as a solution, we are conducting a User Centered Design Research with qualitative research on dissecting beekeepers perspective in order to understand them deeply.

We have found 2 (two) types of rural beekeepers: (1) A traditional beekeepers who believes that his traditional methods are the best, despite any new methods that might be better. (2) An experienced new generation of beekeepers who use inherited old methods but also willing to adopt any new methods possible. In order to create new tools to help beekeeping activities through SAMS, this identification becomes important for implementation strategy within all types of beekeepers in West Java, since there's no such research exist.

[P.06.119] Beekeeping for inclusive rural development: lessons from an up-scaling project in Ethiopia

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We report on lessons learnt in the implementation and adaptive management of various technical, policy and logistic challenges faced by a

commercial beekeeping development project in Ethiopia. This Young Entrepreneurs in Silk and Honey (YESH) project set out to support a total of 10,000 youth in beekeeping and 2,500 others in sericulture with the aim of demonstrating an environmentally and socially inclusive approach to addressing pervasive youth unemployment. Over the first three years of its operations, the project has successfully recruited, trained, equipped and supported a total of 6,447 youth (31% female) in beekeeping, and organized them in to 573 youth beekeepers' enterprises. Once suitable land around developing watersheds and rehabilitating landscapes is allocated by local authorities for establishing apiaries, the project provides series of theoretical and practical trainings in entrepreneurship and beekeeping technical skills development to beneficiary youth recruited based on interest, education, age and gender criteria. Beekeeper's starter kits are then provided ahead of the honey flow season, followed by support to start complementary side-line income generating activities, such as vegetable farming, multipurpose tree nurseries, hive product aggregation, and marketing. Progress monitoring surveys and mid-term evaluation of the project revealed that, apart from skills and knowledge gained during targeted training workshops, prior experience in beekeeping was noted as useful for business success. Timely supply of bee colonies and income generated during the first season were identified as critical for sustaining interest of the beneficiary youth in this new venture. The expected delivery of micro-finance services to the youth groups was constrained by collateral requirements of lending institutions. Effective apiaries and their bee forage nurseries have contributed to strengthening of local efforts to rehabilitate degraded landscapes, also through enhanced societal benefits of job creation to the youth with limited other employment opportunities. This model of setting up commercial beekeeping enterprises in developing watersheds and rehabilitating and degraded landscapes, such as forests, can be replicated in many other countries of Africa where such fragile landscapes need to be rehabilitated but which lack inclusive management plans that take account of direct and indirect benefits to the surrounding rural communities.

[P.06.120] The role of beekeeping in achieving food security and supporting the United Nations' Sustainable Development Goals

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This paper is articulated in two separate level of analysis; the first is to highlight the contribution of beekeeping to food security whereas the second aims at showing the connection and the potential of beekeeping in achieving the United Nations' Sustainable Development Goals (SDGs) that address issues of hunger, poverty, nutrition, environment, climate and several other aspects that are intimately connected with food security.

It will first analyse the four pillars that constitute the dimensions of food security as devised by FAO, namely availability, access, utilisation and stability and it will then illustrate the specific features of each pillar to explain how it operates in practice.

More specifically:

•availability refers to the conditions that enable people to have safe and nutritious food at disposal for consumption;
•access attains to the specific infrastructure and channels that allow people to procure food;
•utilisation relates to the metabolisation of food that it is consumed; and
•stability represents an overarching condition encompassing all the previous three to ensure that they are present and effective over time.

Food security is reached when all four dimensions are present at the same time.

For each dimension, linkages will then be shown of how beekeeping can assist the determination of the conditions leading to establishing and consolidating each pillar.

Having clarified the foundation of food security and the benefits it could receive from beekeeping the paper will look at how beekeeping can assist in the achievement of the UN SDGs.

17 different goals have been identified by the UN to ensure that mankind and especially the more disadvantaged strata of the population particularly in rural areas can overcome crucial livelihood problems and hardships.

A matrix has been elaborated by Apimondia showing the linkages and how different aspects of beekeeping can actually tackle each goal.

The conclusions will show that beekeeping is key in assisting the complex strategies and measures adopted to address the problems identified by the United Nations.

It may serve as the reference for determining how each country on the basis of their specific conditions, circumstances and infrastructure may tackle some of these challenges through beekeeping.

[P.06.121] A model training program to include women and youth to the beekeeping sector living in rural areas of turkey

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Beekeeping is a socio-economic activity in Turkey that has been continuing as a tradition since many years. Turkey is among the most important countries in terms of honey production in the world with its 7.8 million beehives and 105.000 tons of annual crop. However, the average honey production is around 14.5 kg/hive which is far below the world average of 20 kg/hive. One of the most important problems of beekeeping in Turkey is that technical beekeeping practices are not well known because of insufficiently trained beekeepers.

Due to these characteristics, beekeeping is an important source for people who live in rural areas and can be done by all family members. Therefore it is an important business area for women and young people living in these regions.

Within the scope of this program, a literature research has been carried out to determine the status and needs of the target beekeepers. During the study , 455 beekeepers were interviewed for obtaining the demographic structure of Turkish beekeepers.
The Project has been undertaken with the support of "European Bank of Reconstruction and Development" (EBRD), and primarily women and young people living in rural development regions are targeted to be trained as beekeepers. For this purpose, different training programs are organized in the form of on-line learning, utilizing a moto caravan designed for educational purposes and on-site class trainings by working in colloboration with expert academicians.
In order to provide access to the on-line learning program, an accessible web page is designed for all trainees.
For mobile training an appropriate training vehicle is designed and prepared to perform applied on-site training in the field.
Good beekeeping practices, general problems encountered by beekeepers and specific topics requested by beekeepers were chosen as main topics of class trainings. With the establishment of a sample apiary, it is ensured to demonstrate the good beekeeping practices to the newly trained beekeepers.
Eventually different training opportunities will be provided in accordance with the needs of the beekeepers in order to make the best use of the beekeeping potential of Turkey and to ensure its sustainability.

[P.06.122] Low cost materials and simple methods for constructing standard, appropriate and affordable beehives - the case of reducing cost of beehives in the developing countries

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There is positive correlation between the prices of wood and beehives and this is reducing the start-up and expansion of beekeeping ventures in Africa. To avert this situation an innovative beehive production process at a reasonable low cost is being sought
RESEARCH OBJECTIVE
The purpose of the study is to identify the operational ability of selected materials for use in low cost beehive construction.
MATERIALS AND METHODS
MATERIALS
2x2 inches (5x5cm) wooden standard beehive frameworka
Half inch (1.25 cm) U- channel standard beehive metal frame work (knockdown and fixed)
List of hive framework covering materials
aluminum sheets lined with cardboard
wood- less materials eg.
palm Oil branches
woven basket
paint/clay and cow dung mixture painted jute sack ,
husk of coconut fiber boards
frames with the above materials fixed on its openings,
clay tiles
mesh basket
box of cardboard
METHODS
A framework with standard beehive dimensions are constructed, having six opening spaces in a cuboid- like nature by either wood or U-shaped metals.
The top openings are occupied by the top bars and frames respectively whilst the other openings are covered by the above mentioned bee hive framework covering materials. With the metal framework, the materials are fixed unto a wooden frame and inserted into the framework to provide the covering of the opening spaces. However, for the wooden framework hives, they are nailed directly to the wood. The only exception are the elephant grass and oil palm branches which are arranged in a mat-like fashion in a metal holder fixed to the wood.
RESULTS
The Colonized hives means the bees are comfortable with the prevailing conditions in those hives hence could be used to enhance beekeeping amongst the rural folks.
CONCLUSION AND RECOMMENDATION
The principle of the use of various materials both as individuals and in combination with others is based on experiential observation of the nature of materials that harbour the natural nesting sites or crevices occupied by bee colonies hence the recommendation for their use.

[P.06.123] Analysis of technical efficiency and profitability of beekeeping in Adamawa State, Nigeria

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This study assessed apiculture enterprise to determine the technical efficiency and profitability in Adamawa State, Nigeria. A total of four

agricultural zones out of six, and 108 apiarists were selected through purposive and simple random methods, respectively. Descriptive statistics, stochastic frontier production function and budgetary technique were used in the analyses of data. Findings revealed that all (100%) the respondents were males within the age range of 21-30 years (37.9%). While majority (87.0%) of the apiarists were married with household size of 1-5 persons (50.9%), most (50.9%) of them had secondary school education. A larger proportion (37.0%) had between 6 and 10 years of beekeeping experience with 41.6% earning monthly income of N11, 00-N20, 000 from other sources. Further, the findings revealed that the apiarists were technically efficient with a mean of 89.9%, while the inefficiency estimate was only 10.1%. In terms of profitability, beekeeping in the study area was found to have had a high gross margin of N16, 800.00 and net farm income of N15, 225.97 for every beehive in a cropping season. The major constraints to beekeeping reported in the area included beehive crops theft, high propensity of bees' stings, inadequacy of finance, rampant bush burning and deforestation, among others. It was concluded that beekeeping in the study area was found to be profitable and technically efficient. Improving beekeeping business in the area, among other things, would require its modernisation and involvement of female participants, provision of soft credit facilities and enactment of stringent forestry laws to check unwholesome forestry practices.

[P.06.124] Technology for Rural Development: How Apiary Management Software Can Help Rural Economies Grow

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Anyone, anywhere, can be a beekeeper. Low start-up costs, as well as minimal infrastructure and land requirements, have proven beekeeping to be an ideal tool for rural development efforts across genders, cultures, and countries. However, new beekeepers face many challenges to success. Developing Information and Communication Technologies (ICT) tools can be used to improve beekeepers' success, especially in rural areas where traditional training and educational opportunities are limited.
These ICT tools, coupled with standardized data, collaborative tools and analytics support can be used to help increase hive health, best practices adoption, access to markets and communal knowledge sharing. We show how an open apiary management system can be built and shared with beekeeping clubs, associations and NGOs and used for development.
There are several features these systems could have that will multiply the wisdom and experience of an individual beekeeper by augmenting it to that of a larger group, including:
Usage Score - an assessment of how active participants are in using the software. This can help in encouraging more usage and engagement and alerting mentors to needed support.
Health Score - Each participant should be able to see a health score based on a standardized and validated hive inspection process.
Peer Check - Participants should get a report of how their bees are doing compared to other beekeepers in their geographic region or association.
Time Monitoring - Participants should be able to see how their hives are doing over time, comparing multiple years and seasons.
Mentor Dashboard - Building a mentor dashboard to observe the progress and status of new beekeepers and facilitate needed advice in near real time.
Our team is in the process of building, testing and releasing such a system for the specific purpose of driving rural economic development among beekeepers by using these technologies to bring the collective wisdom of local teams in to bear on helping individuals, especially new beekeepers, be more successful in their beekeeping efforts. The goal is to develop and deploy an open system in partnership with bee clubs, NGOs, Governments and others to help make bees for development more successful.

[P.06.125] Analysis of Opportunity and Challenges for Beekeeping in rural areas of Uganda

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This paper documents the opportunities and challenges of bee keeping in Uganda. The information presented here originated from the review of the country reports on apiculture and the field focus group discussion conducted with the Prince Kimbugwes's Foundations Bee keepers members from Kamira Sub-county Luweero district. On the opportunities, the study discovered a number of opportunities all geared to boost bee keeping in Uganda, and these included;- Uganda is licensed to export bee products to any EU member country , there is no registered diseases hindering bee projects in the country, there low costs of land, it's easy to attain knowledge on bee keeping both locally and internally , the country is blessed with good infrastructure such as good road and Internet is networks , and presence of gender inclusion can also be seen as a blessing to the sector. The challenges reported to affect were the existence of predators such as snakes and other small insects that eat the bees and honey, there is no policy strictly set to regulate bee keeping in Uganda and the escalating poverty levels were seen as challenge to bee-keeping and projects in Uganda. However, it was concluded on that, there is need to support both technically and financially bee projects in Uganda in order to boost on the quality of honey, reduce on the predators and loses, and increasing on income and productivity/profitability levels of the projects.

[P.06.126] Three-layered composite materials for diy beehive construction using plastic mesh basket, cardboard and 0.3 mm aluminium sheets

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The limiting factors of the bee keeping industry are; beekeepers knowledge of the bees, environment, supply of bee colonies as well as the cost and efficiency of the beehive. All things being equal the cost of the beehives if reduced drastically will reduce the cost of beekeeping projects and will encourage individuals and governmental participation as the number of beehives to be provided to trained bee keepers will lead to commercial production of honey as against the current situation of providing about two to five hives per beekeeper which is more of a hobby than a business.
Objective of research:The purpose of this study is to evaluate the acceptability by bees for colonization of the innovative beehives constructed with plastic mesh basket, cardboard and thin aluminum sheets.
Materials and methods: Kenyan top bar shaped plastic mesh basket
Cardboard: Thin aluminum sheets (0.4mm)
Methods: The inside of the Kenyan top bar shaped plastic mesh basket was lined with card boards and the outside with the thin aluminum sheets. The two covers were held together in place by copper wires and riveting nails. The aluminum sheets will prevent rain from getting into the hives whilst the card board provides the insulation for keeping the hive's internal temperature suitable for colonization and bee nesting
Results: The hive constructed was lighter and cost effective than wood, with less time used for its construction and management. The hives were colonized after being mounted indicating the bees are comfortable with the internal environment of the hive.
Conclusion and Recommendation.
Reducing the cost of beekeeping projects in Africa is feasible if the above items are used for beehive construction which will lead to mass adoption of the technology. We therefore recommend it for further research with respect to yield of bee products as against those of the wooden hives.

[P.06.127] A sword-like integrated soil imbedded beehive stand,beehive protector and ant barrier against the beehive 'push down' by wind, cattle ,flood , bee badger and ants entry

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INTRODUCTION
Bee colonies and beehives in service left in the woodlots, forests, woodlands and Savannah resources must be protected against various pest and conditions such as lizard attacks, wind effects, cattle activities and floods. To manage this danger against the beekeeping investment, this research was conducted to improve or modify the beehive stands to provide additional protection for the beehives and their colonies.
RESEARCH OBJECTIVE
To develop a soil imbedded beehive stand that provides stability and protection for the hives and bee colonies
MATERIALS AND DESIGN METHODOLOGY
MATERIALS
A 45 (L) x30 (W) x15 cm (H) plastic or metal basin
A 4 inch (10cm) round pipe both plastic (PVC) or metal
DESIGN METHODOLOGY
About 2 pieces of 45 cm of the pipes are cut and one is imbedded in one basin (metal pipes for metal basins and vice versa). 30cm protrusion of the pipe will be at the base of the basin and 15cm at the top. The metal pipe is wielded to the basin whilst the plastic pipe is attached by either hot metal or adhesive. The ends of the pipes at the base are sealed by either a metal plate or plastic (PVC) cups. Thus the set up becomes sword-like with two handles and knives joined together
RESULTS CONCLUSION, RECOMMENDATIONS
The 30 cm pipe ends are fixed into the ground and water or used engine oil are placed into the basin. The legs of the beehive stands for either group or single hive are placed in the pipes. With the stand's set-up in place, the beehives are tied to the stand's platform by 2mm thick wires or jute ropes, thus with any push by wind cattle or floods, the hives are held in place, likewise with the bee badger the top bars are held together by a strip of two pieces of wood across the top bars and also to the hives body hence it will be difficult to push down the hive or open it. Implementation of this concept allows for the protection of both the beehive and the bee colonies as the beekeeper's investment.

[P.06.128] Africanized bee in French Guiana: How to move on to a competitive beekeeping?

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French Guiana, located in the north of Brazil, is a heaven for bees. The environment is still preserved, and the resources are nearly unlimited. In 1971, Africanized bee arrived from Brazil and modified the beekeeping practices. Indeed, Africanized bee is characterized by a strong aggressiveness, a strong propensity to swarm and an amassing instinct almost nonexistent.
In 2016, the Guyanese beekeepers decided to organize a training plan to improve their skills especially in rearing queens and selection. The work was therefore conducted on two axes: livestock development through training in the production of queens and swarms, then setting up a selection plan with the following objectives: honey production, non-aggressiveness, laziness to swarming, disease resistance. Several selection queens from Europe were introduced and tested in tropical conditions. The colonies the most adapted were chosen for multiplication.
The first inseminated queens were produced in 2018 and some moonshine mating fecundations were done. Today, the Guyanese honeybee's livestock is partially transformed to a non-Africanized bee genetic and the results are clearly visible. The production of the selected colonies is 150% higher than the Africanized colony's and the work on the hives is facilitated by an aggressiveness less important. The objectives are, first, to continue the selection work with the same criteria, and secondly, integrate more and more the criteria of varroas tolerances. Indeed, with varroas resistant honeybee, a development of the organic market could be the future of the Guyanese beekeepers.

[P.06.129] Ghana- designed - made simple tools and innovative processes for rearing african queen bees and colony multiplication based on the established queen rearing principles

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INTRODUCTION
Queen rearing books are mostly on domesticated bees of the developed world as well as the architecture of the Langstroth hive as against African bees which are in their natural wild state in Top Bar Hives. This situation calls for the development of tools and processes for easy rearing of the African honey bee queens.
Research Objective
The research seeks to develop tools and processes to simplify the rearing and colony multiplication of African honey queen bees in relation to the established methods
MATERIALS AND METHODS
Materials
1. Queen rearing frame with queen excluder and horizontally placed wooden strips or vertically placed metal strips with combs are angles of 90- 110 degrees respectively
2. U-shaped meshed top bars of external width 2.5cm, length 35cm , height 2.5cm with u-channel pipe (filled with beeswax and patty) placed under the 'U' at intervals of 2.5cm., will have a 4cm strip of young –larva laden combs with a cut out to have a 2cm finger-like projection downwards to mimic the Miller's method below will be fixed into it
3. U-shaped metal frame-like comb holder system of width 2.5cm, length 35cm and height 22cm to hold whole combs –Hopkins method
4. Unsealed brood comb with hatched larvae of age 1- 2.5 days
5. Beeswax and patty
6. Feeder for both solid and liquid feeds and feeds
7. Inverted triangular shaped combs to be fixed into (2)
Methods
1. Select a Breeder Hive
2. Place empty old combs into the brood sections of the Breeder hive
By the 4th day the eggs might have been hatched into very young larva and the combs will be turned horizontally with the cells facing downwards and placed in the above frames or as follows in a Cell Builder Hive
A. Queen cell grafting into natural or synthetic queen cell cups
B. 'Cubing ' of combs (15x 15mm-3 cells) for queen cells formation and fixing them onto breeding frames by molten wax
C. Strip cutting of young –larva laden combs
RESULTS AND CONCLUSION
Methods are effective and recommended for queen rearing and colony multiplication

[P.06.130] In- hive feeder for solids (patty) and liquids (water and sugar solution) feeds -the case of increasing the feeder's surface area for effective feeding and reduced bee drownings

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INTRODUCTION
Extra feeding of bees are needed during winter, prolong rainy and dry seasons, colony division and queen rearing activities. Feeds could either be in liquids or solids and/or paste forms. Various forms of feeders for dispensing liquid feeds have being developed but not much for the paste or solid forms which are mostly placed on top of the frames in the Langstroth hives. This therefore calls for the development of an innovative in- hive feeder for both feed types where the bees feed at various levels to improve feeding efficiency with much reduced bee drowning.
MATERIALS AND METHODS
MATERIALS
7 Inverted- shaped plate of length 24cm, width 2cm and thickness range 0.8-1.5mm
5 pieces of ¾ in of u-channel pipes of length 40cm
48*3.2*2cm top bar wood with centrally placed groove to fix and hold the feeder in place by u-shaped nails
DESIGN METHODOLOGY
The 5 pieces of the u-channel pipes are wielded and arranged in a storey fashion at equidistant from the base of the 7 inverted-shaped metal plate leaving a space of 2-3cm amongst them to allow the bees to feed comfortably. The set-up is then fixed under the top bar by either u- nails or screws and hanged in the hive to replace a top bar or frame in terms of top bar or Langstroth hives respectively for patty and small volume of liquid feeding to prevent drowning of bees
RESULTS, DISCUSSIONS AND RECOMMENDATIONS
The feeder enables the bees to feed comfortably as compared to placing the patty on top of the Langstroth during winter. The provision of sugar solutions and patty to bees in the top bar hive is not a common practice in Africa as most of the feeders in the system are designed with the Langstroth hive in mind. It can also be used to provide water or sugar solution to the bees without the incidence of drowning as the series of the feeder's cisterns are not deep .The feeder can be applied to both the top bar and Langstroth hives during colony division, queen rearing, winter periods, prolong dry and rainy seasons.

[P.06.131] Pilot project Beekeeping in Bangladesh

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The beekeeping is a great way to establish stabile household income and improving the quality of life for vulnerable social groups. Beekeeping requires little start up investment and does not require complex technologies and techniques to start. Beekeeping therefore offers employment opportunities for youth, women, farmers and large families.
As a result of activities for the World Bee Day Slovenia received Beekeeping Academy of Slovenia (BAS). The BAS was established to improve beekeeping knowledge in international arena, to improve the meaning of bees and wild pollinators, to arise the promotion of apitourism and opportunity for job creation.
The BAS offers informal training courses in beekeeping for instructors, for groups interested in beekeeping education and special programs for developing countries with traveling trainer. Upon its establishment, the BAS has taken over the "Beekeeping in Bangladesh" pilot project. The aim of the pilot project is to develop small-scale family-run beekeeping establishments which will contribute to the household income, improving the quality of life for inhabitants in Bangladesh. Another aim of the project is to raise the level of the whole technology of beekeeping in Bangladesh.
The program consists of on-the-spot analysis of the area and the current state of affairs in beekeeping, a risk analysis of the project implementation and health status of the bees, an analysis of the queen breeding and bee colonies, market analysis and a detailed timeline for the individual project phases.
The main goals of project are: establishing of reference apiaries, queen breeding with improvement of genetic material, educations for the local trainers and market aspect of honey bee products.

[P.06.132] Monitoring the health status of Carniolan honeybee colonies (*Apis mellifera carnica L.*) in Bangladesh

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Twenty Carniolan honeybee colonies were exported from Slovenia to Bangladesh in January 2019. Bee colonies originated from an apiary free from the American foulbrood, European foulbrood, chalkbrood, small hive beetle and Tropilaelaps spp.. Bee colonies were treated with oxalic acid dihydrate for the control of Varroa destructor before nucleus colonies were established and exported as package bees. From the winter cluster honey bees had to cope with new stresses – hot weather and pathogens such as Aethina tumida, Vespa velutina and Tropilaelaps spp. The health status of imported bee colonies was monitored during first half of the year. Diagnostic tools for bee diseases were also used with comparative group of twelve Italian honeybee colonies (*Apis mellifera ligustica*), which were introduced in Bangladesh in the last decade of 20th century. As foreign race in Bangladesh, Carniolan bees are more susceptible to diseases than local races. Due to climate change, knowledge about the response of the European honey bee, *Apis mellifera*, to potential new stressors is very welcome.

[P.06.133] Capturing honey bee colony swarms at the hive's entrance-the case of applying queen capture mechanism at the hive's entrance

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The swarming characteristic of the African bees which was formerly thought as disadvantage is seen now as a means for protecting the colony against diseases and pest infestation. This calls for a technological research to develop an entrance mechanism for swarm capturing as bees are trainable observing from the bee's life activities from the young to the adult stage.
Research objective
To technologically develop a mechanism for capturing bee swarm at the entrance of the beehive
Material and Design methodology
Material
10 pieces of U-channel pipes of length 30cm each
5 pieces of 30 centimeter square Queen Excluder mesh of cut out dimensions of 2.7 by 4.03 cm
One inch angle iron
Half-inch galvanized flat bars
One inch flat bars
Design methodology
Two U-channel pipes were wielded at 90° along their lengths. Thus whilst one's open end is facing the (left) 270° coordinate direction the other is facing (downwards) 180° coordinate. Four pieces of this set was done and for each pair a U-channel pipe slanted at 45° and facing the 270° direction was wielded to join at its ends to form a two Z shaped system. Four flat bars (30cm) were wielded at both the top and bottom to form a box-like system. The mesh are inserted into the U-channel's open ends to cover the box-like system but the front has the slanted mesh so that the bees have to move a distance of about 3cm after passing through the mesh to get into the hive
In un-colonized hive the system is put in place without the front mesh but later it is gradually placed there after colonization whilst in the colonized hive, the system is put in place without the mesh but gradually the mesh is inserted into the non-flight path of the bees then later the flight path gradually.
Results and Conclusion
When bee swarm they assume that because they can go through the mesh the queen can do same but she will be trapped for collection because of her size. Bees are trainable hence they get use to the queen excluder mesh

[P.06.134] How to define the sustainability of bee farming systems?

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Floral resources availability, annual climatic conditions or state of the honey market: professional beekeepers have to cope with several economic and environmental challenges to match their expectations in terms of economic, environmental and social performances. In other words, to ensure the sustainability of their farm. But beyond some apparently consensual goals as a minimal income or a good work-life balance, these expectations vary from a beekeeper to another and the sustainability of bee farms remains undefined. Still, such a definition would be useful to better identify the current issues of professional beekeeping, and to develop a sustainability assessment tool that would be more adapted to beekeeping than the tools that are already used in other agricultural sectors.
Then, how can we define the sustainability of bee farms to include the diversity of beekeepers' expectations and viewpoints? Through a collective work with French professional beekeepers and other stakeholders from the apicultural sector, we adapted this concept of sustainability to the specificities of bee farming systems. Various viewpoints on the sustainability of bee farms were collected through individual interviews with beekeepers, beekeeping advisors, research and development experts and other stakeholders from apicultural sector. The numerous suggestions for sustainability items that resulted from these interviews were then collectively discussed and organised into a consistent framework.
This framework includes technical, economic, social and environmental issues that are distributed into six main themes: economic viability, quality of life, environmental impact, local development, contribution to current stakes of the beekeeping sector and society, and

production security. It emphasizes some specificities of beekeeping compared to other agricultural sectors and provides an overview of the main current issues of professional beekeeping. This sustainability framework is the first step to develop an assessment tool at farm level. It could be used to communicate about current issues of professional beekeeping, to identify strengths and weaknesses of a bee farm or to analyse and discuss the sustainability of a farm establishment project.

[P.06.135] The SAMS Project: International Partnership on Innovation in Smart Apiculture Management Services

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SAMS is a project funded by the European Union within the H2020-ICT-39-2016-2017 call. SAMS enhances international cooperation of ICT (Information and Communication Technologies) and sustainable agriculture between EU and developing countries in pursuit of the EU commitment to the UN Sustainable Development Goal “End hunger, achieve food security and improved nutrition and promote sustainable agriculture”. The project consortium comprises four partners from Europe (two from Germany, Austria and Latvia) and two partners each from Ethiopia and Indonesia. Beekeeping with small-scale operations provides perfect innovation labs for demonstration and dissemination of cheap and easy-to-use open source ICT applications in developing countries. SAMS allows active monitoring and remote sensing of bee colonies and beekeeping by developing appropriate ICT solutions supporting management of bee health and bee productivity and a role model for effective international cooperation. SAMS addresses requirements of end-user communities on beekeeping in developing countries. It includes technological improvements and adaptation as well as innovative services creation in apiculture based on advanced ICT and remote sensing technologies. SAMS increases production of bee products, creates jobs (particularly youths/women), triggers investments, and establishes knowledge exchange through networks. To find out more visit our project website https://sams-project.eu/.

[P.06.136] Beekeeping potential and the utilization in the red dwarf honeybees, *Apis florea*

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Apis florea is a honeybee species which is widely distributed through the warm climate of Asia. It usually builds a single comb approximately 20 – 30 cm width in shrubs, bushes and small trees. This bee is highly adapted to build their nests in many kinds of habitats such as disturbed areas, urban areas, intensive agricultural areas, and savanna ecotopes.This study suggested beekeeping with A. florea can be developed for sustainable beekeeping particularly for Thailand. This research projects aims 1) to investigate *Apis florea* bee flora; 2) to study comb developmental cycle of them; 3) to develop a low cost equipments to facilitated A. florea beekeeping; 4) to develop technique producing honey 5) to compare the cost effectiveness of developed method and conventional honey hunting method. The total yield of honey from beekeeping and natural honey hunting was 1,229.56 ± 230.26 g and 270 ± 89.92 respectively. The beekeeping technique developed in this project yield 4.5 time higher than honey collected from natural honey by the conventional hunting method. There was a distinctly difference between the cost per year gained for conventional honey hunting than developed beekeeping technique. In the same investment of timing, conventional honey hunting lose 16,743.8 bath per year whereas developed beekeeping technique gained 57,445.61 bath per year. In conclusion, *Apis florea* is highly recommended potential for economically beekeeping.

[P.06.137] Beekeeping in Canada: Assessing Trends of Self-Sufficiency and Participation in the Global Honey Bee Trade

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This thesis examines trends in beekeeper self-sufficiency and commodification of honey bees to better understand the barriers and pathways to sustainable and healthy economic and socio-ecological beekeeping regimes. The Canadian beekeeping industry depends on external sources of honey bees to maintain current populations, however the benefits of locally-raised bees is widely acknowledged. Canada employs two equally common approaches: i) use of imported bees, and ii) domestic and individual self-sufficiency. Notably, a mixed-model is developing that propagates Canadian bee stock externally and imports it to Canada in early spring. Well-established Canadian beekeeping regimes, with geographically specific sets of productive and organizational patterns, have been slow to increase domestic self-sufficiency. Common explanations for this focus on environmental constraints of Canada’s northern locations that prohibit economically feasible self-sufficient commercial beekeeping. However, this study shows that rather than only environmental, biologic, and economic constraints to beekeeper self-sufficiency there are also significant social barriers entrenched in beekeeping cultures. This study highlights, in particular, how the politics of hive management are shaped by choices of participating in the global bee trade or beekeeping with rootedness in time and place; resulting in linear or integrated on-the-ground beekeeping practices. This research demonstrates the social and material practices that contribute to creating robust and desirable beekeeping regimes, specifically, how hive management is influenced and how social networks and institutions develop and promote beekeeping ideologies. Creation of regional and national beekeeping networks and institutions has been key to building the industry and (re)developing the required local knowledge for self-sufficient regimes. Some positive impacts of beekeeper self-sufficiency were found to include, creating deeper ecological connections,

deepening bee and local knowledge, minimizing vulnerabilities inherent in the global lively commodity market, and potential roles to play in creating unique approaches for national biosecurity and challenging the structure of our current agri-food systems. Understanding the trends and value of domestic and individual beekeeper self-sufficiency and the commodification of honey bees is essential to re-imagining more democratic economies and embracing the potential to reshape beekeeping cultures and industry in ways that are economically and ecologically sustainable, especially in times of precarious markets and changing climate.

[P.06.138] How to reduce hunger in the world and improve the ecosystem with beekeeping and the use of beebread collector

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Climate change is the direct consequence of global warming. The increase in carbon dioxide, pollution, intensive farming, deforestation, etc. are progressively increasing the temperature of the atmosphere causing floods, melting of the polar ice caps, rising seas,, drought, etc.
In particular, the prolonged absence of rains favors the desertification of vast territories, increasing the food shortages of resident populations. Today more than 800 million people suffer from hunger and about 400,000 people die of malnutrition every year. The poorest populations are affected, in particular children living in developing countries. Pending a solution to the problem, the spread of rational beekeeping in poor countries, combined with a specific agricultural program, can help integrate the diet of hungry populations by providing carbohydrates with honey and proteins with bee bread. Until today it was not possible to use bee bread without destroying the combs. The invention of the beebread collector offers the possibility to extract the bee bread contained in the cells of the honeycombs without damaging them. In this way the honeycombs, after having been emptied, can be inserted again in the hives avoiding a laborious reconstruction work to the bees. For this project only honeycombs that can be extracted from the hive can be used (Langstroth, Dadant, top bar hive, etc.).
Bee bread can enrich the diet of adults and children and, due to its high protein content, replace meat and eggs. In fact 100 gr of bee bread are equivalent to 500 gr. of meat or 7 eggs. Depending on the climatic and environmental conditions from a beehive, during the year it is possible to extract 200 to 1000gr of bee bread. To produce 1Kg of meat, thousands of liters of water are needed, soil exploitation, the use of fertilizers, and the administration of insecticides. Beekeeping combined with a rational, non-invasive agricultural program can provide poor populations with high quality proteins, drastically limiting water consumption and soil, helping to contain desertification.

[P.06.139] Data survey on the socioeconomic, environmental and technological situation of beekeepers in the state of Espírito Santo, Brazil

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Beekeeping may be developed by small producers with significant return and low environmental impacts (SCHOWALTER, 2000). In this sense, the activity should be highlighted in the current scenario of environmental degradation and devaluation of family farming. The work presents a data survey on the socioeconomic, environmental and technological profile of beneficiaries of the APISFRUT program, an alternative of the Federal Government for the productive inclusion of socially vulnerable families in the state of Espírito Santo, Brazil. Overall, 517 producers from 24 municipalities took part in the study, from which 264 answered to a semi-structured questionnaire in order to obtain data related to their profile and perception of beekeepers.
Regarding scholar degree, 27% of the interviewed people had finished high school and only 4% had concluded the under graduation. Family farming was predominant: 40.1% of the answers indicated that only one person of the family was involved with the activity, while for 22.9% two people were involved.
Regarding economic aspects, beekeeping was indicated as the main activity by 13.7% of the beneficiaries, as complementary activity by 82.4% and as hobby by 0.8%. In relation to financial control, 76.7% of the beneficiaries did not record the production costs. In fact, only 6% showed to be aware of the costs, while 9.9% knew partially.
Environmental issues are crucial for the development of beekeeping, since they influence the health of swarms, and consequently their capacity to pollinize and produce. Considering a radius of 3 Km around the apiary, the main crops were coffee (57%) and eucalypt (27.5%), which were also responsible by the main flowerings, with 39.7% and 29.7%, respectively, followed by the wild flowering, with 11.5%.
Technological aspects included the participation in training courses and use of technologies. Data demonstrated that 64% did not take part in trainings in the last two years and 82.8% of producers did not use informatics in the activity management.
The current study contributes with data for APISFRUT to promote beekeeping not only as a mere complementary income, but as a promising entrepreneurship that may strengthen family farming and then improve socioeconomic, technological and environmental issues.

[P.06.140] Preferential loans for the development of beekeeping in Samarkand

O. Khaydarov, K. Rashidov
Samarkand Region Beekeepers Association, Samarkand, UZBEKISTAN

Preferential loans for the development of beekeeping in Samarkand.
The National Exhibition Complex of Uzexpocenter on December from 1 to 10 of the current year hosts the honey festival and the exhibition-fair of honey products. More than 250 subjects of beekeeping from all regions of our country participate in it with its products. As part of this event, the regular session of the International Press Club was held.
At the meeting, under the motto of Uzbekistan Beekeepers: New Direction, New Reforms, Chairperson of the Association Association of Beekeepers of Uzbekistan Sh. Suyarkulov and specialists of the sphere spoke.
As noted, the decree of the President of our country Shavkat Mirziyoyev about On measures for the further development of the beekeeping industry in the republic dated October 16, 2017 is an important factor in the development of the sphere at a new stage.
Currently, there are about 13 thousand beekeeping subjects in Uzbekistan. This year, the beekeepers of our country have received 15 thousand tons of honey and honey products. It is assumed that by 2021 this figure will exceed 23 thousand tons. For this, the necessary conditions are created, new technologies are attracted to the sphere, scientific research continues. In particular, the association of Uzbekistan Beekeepers implements measures for the systematic organization of technological processes for the production of honey products, the cultivation of packages of bees and the strengthening of the food supply base, the rational use of natural resources.
So, we have to expand the production of high quality honey, said that Sh. Suyarulov, the chairman of the Association of Uzbekistan Beekeepers. For this, first of all, it is necessary to organize training. We are planning to organize beekeeping directions in agricultural universities.
The event also talked about the plans for the future of the association Uzbekistan Beekeepers. The current tasks of the association are to join the International Federation of Beekeeper Associations in Apimondia, to provide legal and financial assistance to beekeepers, to further increase export volumes.

[P.06.141] Effects of Honey Bee comb chaff on water logged soil (Fadama), growth and yield of Amaranthus hybridus in Akure South West, Nigeria

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The experiment to determine the effects of honey Bee comb chaff (HBCC) on Fadama soil was conducted at the teaching and research farm of the Federal University of Technology Akure (FUTA), Ondo State. The site lies between longitudes 7oE and 7o45'E and latitudes 6oN and 7oN of the equator. Composite soil sample were collected from the sites before the experiment and a sample per plot at the end of the experiment. Common Amaranth hybridus seeds were obtained from the National Horticultural Research Institute (NIHORT) Ibadan, Oyo State. The treatments consisted of three levels of HBCC: 0 t ha-1, 10 t ha-1 and 20 t ha-1 replicated 3 times. Honey Bee comb chaff was obtained at Hope Agricultural consult Akure. The HBCC was applied by working it into the surface of water logged soil and allowing it to cure for four weeks before planting the A. hybridus seed. Seeds were planted at spacing of 30 cm x 10 cm on beds measuring 2m x 2m; seedlings were subsequently thinned down to one seedling per stand at 2WAP. Soil temperature and moisture were monitored. Soil samples collected were taken to the laboratory, air dried and analysed for Soil bulk density, % sand, silt and clay, moisture content, pH, Organic carbon, Nitrogen, Phosphorus, Calcium, Magnesium, Potassium and Sodium. Data were also collected on A. hybridus growth parameters: number of leaves, Crude Leaf Area (Length and Width) (cm2), harvested fruit length (cm), girth (cm) and weight (g). All HBCC combination improves considerably the structure of the soil and significantly high in organic carbon content. Addition of honey bee chaff 20 t ha-1 was found to have improved soil bulk density, porosity. O.M. N, P, K, Na, Ca, Mg, ECEC and A. hybridus yield parameters. 20 t/ha HBCC gave the highest vigor, growth and yield though not significantly different from 10 t ha-1 HBCC at $P < 0.05$.

[P.06.142] Beekeeping survey in all over Afghanistan for the year 2019

G.S. Satti
FCO, Kabul, AFGHANISTAN

Beekeeping in Afghanistan provides an attractive income generating opportunity for poor families and small landowners, as its investments are limited and the market for honey is growing. The Afghan bee is very gentle, which makes it easier to handle. The Afghan flora offers a wide range of high quality honeys, which makes Afghanistan an exceptionally good region for beekeeping.

POSTER SESSION 12 08:30-18:00
BEEKEEPING FOR RURAL DEVELOPMENT II POSTER AREA

[P.12.294] Which problems are faced by bee keepers and bees

N. Singh
Pau Kissan Club, Ludhana, INDIA

Safe bee keeping

[P.12.295] Honey Granulation: A new challenge to beekeepers in Southwest, Nigeria

A. Akande
Jorafarm and Associates, Osun, NIGERIA

Recently the campaign for honey is on the high side in Nigeria because of its medicinal importance and cultural uses among Nigeria. Based on the percentage of fructose and moisture content, honey can either be harvested as granulated or non-granulated. However, buyers prefer to buy non-granulated honey as they perceive granulated honey has been adulterated. This portends fear to many local beekeepers because they have never experienced such in the past until lately. We carried out our studies in Osun and Oyo states, Nigeria among the beekeepers who experienced honey granulation due to change in weather between 2015/2016 and 2018/2019. In addition, few local beekeepers who were around during our southwest beekeeper forum quarterly meeting with the participation of the following states (Osun, Oyo, Ogun, Kwara, Ondo, and Ekiti) also shared their experiences. Fifty participants were selected, and questionnaires were administered. The results showed that granulation decreased the market value of honey because buyers believed that the local beekeepers had added sugar, which eventually caused the honey to granulate. During this period of change in climate condition over 85% of the honey harvested were crystallized. In contrast, less than 10% of honey harvested in 2016/2017, 2017/2018 were granulated.

BEEKEEPING FOR RURAL DEVELOPMENT

12 SEPTEMBER 2019

POSTER SESSION 21 08:30-18:00
BEEKEEPING FOR RURAL DEVELOPMENT III POSTER AREA

[P.21.539] A Queen for the People: A comparative study of Walk-Away Split Queen Bees verses Grafted Queen Bees

S. Comfort
Anarchy Apiaries, Hudson, NY, USA

While it is generally believed that bees will rush to make a lower quality queen from an older larvae in an emergency queenless situation, there have been studies (Tarpy et al.) that suggest worker bees have a conscious quality control and will terminate inferior queencells. By submitting both grafted queens and bees' choice queens to Tarpy's Queen and Disease Clinic at NC State University for morphometric and spermatheca analysis, I have accrued some data showing that in fact bees know best- queens that worker bees choose to raise are often superior quality. I will review graftless queen production methods, various ways of inducing a nucleus colony to raise a quality queen, and discuss walk-away split methods from three commercial operations: Anarchy Apiaries (Sam Comfort), Champlain Valley Apiaries (Chaz Mraz), and Lusby Apiaries (Dee Lusby). The technicalities of raising queens by grafting into cellraiser colonies can be intimidating. Knowing that simpler methods are viable on a large scale could help many beekeepers grow their operations.

[P.21.540] Mitigating 'Park-People Conflict' Using Beekeeping to Control Elephant's Raid on People's Crops: A Case Study of Communities Adjacent to Kibale National Park in Western Uganda

R. Ndyabarema
Kabecos, Kampala, UGANDA

Kibale forest National Park in Western Uganda established in 1993 has been the scene of conflicts between local communities and wildlife managers. Population growth and scramble for land has made wildlife share the available space and resources with people. This often result into Park–people conflict ranging from wild life managers apprehending local people accused of illegally collecting non-timber forest products, to serious confrontation with poachers and arrests that occasionally result in deaths of both people and wildlife. On the other hand wild animals whose movement is not curtailed by park boundaries roam the entire landscape including human settled areas and farmland leading to damage of crops. This has been one of the main causes people-park conflict around Kibale National Park.The animals that destroy people's gardens (Elephants) are of conservation importance, but people perceive these as problem animals that impoverish their livelihoods. The solution to crop raiding has been found and this is anticipated to solve people –park problem.

This paper therefore outlines an approach undertaken by Kamwenge Beekeepers cooperatives (KABECOS) through its partnership with Uganda Wildlife Authority to deal with the problem of Park- People conflict, while conserving elephant populations. It involves the siting of beehives along the park boundary as a deterrent to crop raids by elephants on peoples farmlands.

The paper synthesizes the project indicators reducing People-Park Conflict
Reduced incidences of wildlife crop raiding
Enhanced household incomes through Honey sales
Reduced number of community members arrested due illegal access
Absence of bush fires.

The paper crystallizes the impact this project has made on the community livelihoods including enabling the communities to harvest their entire crops without disturbance, harvesting honey and other products and protecting elephants from poachers. The benefitting communities, have become key agents of UWA, hence sustaining park conservation.

In conclusion; this project has improved tolerance and co-existence between UWA and communities in the parishes adjacent the National park hence reducing people-park conflict in the project area. It has enhanced wildlife management and community development leading to improved livelihoods of park fringe communities while seeking to attain the objectives of wildlife conservation.

[P.21.541] Beekeeping in the Sudan

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Since the 1920s there have been attempts to handle honey bees in Sudan in a more scientific and economic ways. For several reasons the success was insignificant. The agriculturists are quite aware of the potentialities of honey bees as producers of well rewarded honey, wax and as pollinators. *Apis mellifera* was originally reported in whole Africa. There are a lot of strains and bee races. The mitochondrial DNA analyses showed that all wild honey bees exclusively belonged to African haplotypes. However In some studies nonnative haplotypes were revealed in managed colonies on apiaries reflecting unambiguous evidence of imports from European stock. It was suggested that variation in polyandry in wild honeybee populations of Sudan is primarily driven by climatic differences among ecosystems variation rather

than by the absolute or effective local honeybee population size. Today 80% of bee honey is obtained from tree cavities and caves in the mountains. As a way of advancement some of the natives constructed holed tree trunks, sorghum stalks and bamboo splits for the bees to occupy naturally. In 1930 the government entomologist imported European honeybee race in Langstroth hives from England which failed to adapt. In 1965, successful apiary for *Apis mellifera* race in Langstroth hives was established at Kenana Research Station. The 1976 a survey for bees resulted in yellow, black and mixed strains. Bee culture should be promoted at southern Darfur, southern Nuba mountains, the Blue Nile and Central Sudan.In addition to *Apis mellifera*, in 1985 the dwarf Asian bee Apis florae was discovered in Khartoum. The future of beekeeping in the Sudan depends on the local strain of *Apis mellifera*. Honeybee swarms can build up strong colonies if kept in appropriate hives and managed adequately. Only knowledgeable and interested people will succeed in promoting bee culture in the Sudan.

[P.21.542] Queen Rearing in the Sustainable Apiary

M. Palmer
French Hill Apiaries, Saint Albans, Vermontt, USA

It wasn't long after starting my nuc wintering program, that I began raising my own queens. I got tired of buying hundred packs of mated queens, only to find half of them to be rubbish. Although I had never really raised any queen bees, at least not seriously, I felt that if the queens I raised were half rubbish, at least they were free. This decision has served me very well.
In my opinion, a beekeeper can raise better queens than can be bought on the open market. Breeding stock is selected from the apiarist's best stocks, being maintained under the apiarist's management, and in the apiarist's microclimate. But the problem arises. How does one raise a large number of quality queens in an economical and sustainable way? As a honey producer, is it wise for me to sacrifice part of my honey crop by devoting an apiary to cell building?
The answer, once again, is the nucleus colony.
By allowing overwintered nucleus colonies to build up in the spring, I can harvest brood from them to be used where needed in the apiary. In 2011, from 50 of these "Brood Factories", I harvested enough emerging brood to create 35 powerful cell builders that, over a month and a half, gave me more than twelve hundred queen cells. The factories also gave me enough additional brood to create 330 nucleus colonies, to be wintered over with the new queens.
Over the years, the power of the nucleus colony has defined my life's work. It has enabled me to create a breeding program in my apiary that has me sending quality stock across North America, north and south of our borders.
I believe I have a solid plan for selecting good breeding stock, and nucleus colonies have become an important part of evaluating that stock during the first season.
I hope to share my work in breeding honey bees, in a sustainable way, over the last 20 years, along the Quebec/Vermont/New York borders

[P.21.543] Adapting to Parasites with a Sustainable Apiary

M. Palmer
French Hill Apiaries, Saint albans, Vermont, USA

I began keeping bees in 1974, when things were much simpler and cheaper. I paid $10.50 for my first package bees. Once I learned the beekeeping essentials, it wasn't difficult to maintain my colony numbers. The only real danger we faced was American Foulbrood. While deadly to honey bee colonies, the disease was easily prevented and controlled. By the end of the 1980's, I had built my operation into an apiary of nearly 1,000 colonies, pollinating apple orchards with 600 colonies. Everything seemed nearly perfect.
Then we experienced new parasites in rapid succession and beekeepers had to adapt or close up shop. First, we adapted our beekeeping to the tracheal mite, Acarapis woodi, and then the parasitic mite, Varroa destructor. Beekeeping costs increased and beekeeping management became more complex. I had a difficult time keeping my bees alive and fulfilling my pollination contract. Losses forced me to buy singles from South Carolina to replace my dead-outs. At $55 each, today they would be considered affordable. But in the 1990s my contract only paid $30 per colony. Losing $25 per colony to fulfill my contract was not a good business plan.
I was forced to become self-sustainable. In 1998, I began overwintering nucleus colonies, made up during the main summer flow. I am based in upstate Vermont and New York, near the Canadian border. Once my sustainable apiary management system was perfected, I no longer had to buy bees. My Canadian counterparts are now in the same place I was back in the 90s. They're forced to buy expensive bees to replace dead-outs, so they can fulfill their pollination contracts. They're spending top dollar to purchase Australian package bees, to fulfill a pollination contract that doesn't recover the cost of the bees. Such importation puts North America at risk of importing a new mite, Tropilaelaps clareae. Using my sustainable system, nucleus colonies can be wintered anywhere in Canada where colonies traditionally survived the winter. I will discuss how to sustainably build and overwinter nucs in northern climates, so commercial beekeepers can become self-sustainable.

[P.21.544] Beekeeping for the conservation of watersheds in Peru

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In the San Martin region, since 2009, an initiative grew to intervene the conservation of areas in the watershed due to high deforestation

that was taking place at that time.
In order to mitigate the environmental impact, a project called, Recovery of systemic eco services in an area of high environmental value in Moyobamba, arose with the support of the regional government. The main goal of this project was to strengthen beekeeping activities in the region. In 2016, there were 15 beekeepers who ran 450 beehives, and in 2017 they won first and third place at the Peruvian national contest of quality and innocuousness of honey.
Alto Mayo is a valley rich in flora and fauna. It has a subtropical climate of 25°C on average, a rainfall median of 1354 mm per year, and a relative humidity between 76-95%, in an altitude from 750 to 1200 m.a.s.l. Bloom curves in Alto Mayo have their peak in the months of July, August, September and October. These conditions enable the development of an environment for beekeeping activities. In 2107, the production areas, which are secondary forests, yielded 30 kgs. per hive; they thrived because of the clean waters and remoteness from pollution of single crops. The harvesting was done by removing the beehives with more than 90% of the frame cells capped, just one month after the bees deposited the nectar into the honey super box. After extraction, honey with 17% humidity and 83 brix was obtained.

[P.21.545] Diversity of Savannah tree species are key to bee health

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The diversity of natural diets is key to bee health and most of the savannah tree spp bees forages on are medicinal. Beekeeping in Northern Uganda with diversity of tree and grass species like Terminalia , Combretum , Africana doniana, Vitallaria paradox, Rufia spp and many other are bee forages. They are the best for sustainable and natural beekeeping for rural development. It makes African bees very unique and active and resistant to Varroa Mites and other bee diseases as well which is a big problem in commercial beekeeping in developed countries. In natural beekeeping feeding of bees is limited. Different plant species flowers at different times of the year and honey can be harvested throughout the year with three main harvesting season, March, June and October.
Beekeeping for starter is best with traditional hives like log hives and woven basket hives since the material are easily assessable from the local environment, it is cheap and bees rarely abscond once colonized even if left in the bush un- attended to. Colonies become stable and strong to defend themselves.
Community benefits of sustainable and natural beekeeping is enormous because their levels of participation in hives technology, value addition, siting, harvesting and processing is wonderful.
The existence of small medium enterprises like, Hive Uganda, Gulu Natural Honey, Gulu Beekeepers Training and Resource Centre, World of Bees, Koch Goma Beekeepers Association, Tropical Honey, Malaika Honey, and the Local Governments are some of the energizers in the realization of this dream. Regional cooperatives to empower the bee farmer for commercialization to boost honey productivity for export is in the offing but linkage with international partners for beekeeping inputs, skilling and value addition for exports could results to speedy realization of the goal.

[P.21.546] Sustainable Management of Beekeeping in Nepal

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It has been learned from previous development efforts that the conventional development models were more of top-down and centrally driven and no longer effective and sustainable. These development efforts were failed in most cases due to the fact that very little attention was given in responding to the real needs of beneficiaries. The development efforts had ignored the role of local organizations in achieving sustainable development.

[P.21.547] Beekeeping in Afghanistan (Nangarhar Beekeeprs Asssociation)

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Afghanistan has good climatic condition for all agricultural and animal husbandry rearing and production, especially for beekeeping. Afghanistan is covered by snowy mountains and various green valleys, therefore there are more than 2000 small and medium beekeeping farms and from 5 to 6 labor are involved in every beekeeping farms and only more than 900 beekeeping farms are exist in eastern region of Afghanistan because the Eastern region is also known as Ever Green region of Afghanistan. Therefore, eastern region has always the flowers season in different areas at different times. The season flowers are the reason of beekeepers attraction in this zone and all Afghanistan beekeepers bring honey bees to eastern region and get their production in eastern region.
Beekeeping is one of the traditional and profitable agri-businesses and there are many people are involved in this business which generates sufficient income to feed their families.
People involved in this business have many challenges including technical deficiency, and lack of markets.
Beekeepers in Afghanistan generate sufficient income to feed their families. So by cultivating berry, orange, Malta orchards that more than 11000 poor people will involve in this business and more than 15000 gardeners and beekeeper farmers will get benefits from these trees.

Create 2000 new beekeeper farms it will increase the yield of agricultural crops due to the contribution of honeybees in pollination process and maintaining of ecological balance and from other side many jobless people will involve in beekeeping business.
As the fighting are finished in the country, the rehabilitation of country are in progress ,there is good chance and opportunity to develop and strengthen the honey bee industry ,this well help in the development of people economics and people well get resistance against the disease. So there is need for government, national and international agencies to work for this sector, to solve the problems and hardships of honey beekeepers and honey businessmen and make some facilities for them.

[P.21.548] Chalkbrood disease in vietnam: epidemic characteristics and control method

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Vietnam has about 3,000 migratory professional *Apis mellifera* beekeepers, managing 800,000 colonies and producing 50,000 tons of honey per year. Like *A. mellifera* colonies in many other countries, those in Vietnam have been facing with many bee diseases and parasites including the mites Varroa destructor and Tropilaelaps sp. However, with the effective bio-technical control methods, the beekeepers can keep colonies strong and produce honey without miticide residues.
Chalkbrood is a disease which is caused by the fungi Ascosphera apis and Ascosphera alvei. It has been known for a long time by beekeepers in Europe, America, Australia and many Asian countries but it has never been seen in Vietnam until a few years ago. However, by somehow the disease invaded into the country and it has been now causing damage to the beekeeping industry.
A study was carried out together with beekeepers in the field during 2015-2018. It is aimed to find out a method to control the new disease effectively but safely for honey to export and economically for the beekeepers.
Our study results show some of the epidemic characteristics of Chalkbrood and find out a bio-technical method which is rather effective for control of the disease, safe for the export honey, economic for the beekeepers, and sustainable for the beekeeping industry.

[P.21.549] Rural development of beekeeping

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I will give information about beekeeping,specially for natural beeking having no need of pesticides and pets.
Next,about bees are working as a pollinator to Increase the production of various crops

[P.21.550] Recovery of agroforestry ecosystems in protected natural space “siete lomas”. Integrating the beekeeper as a collaborator

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Multiple studies show that sustainable agricultural development favors the physical process of healthy ecosystems, biodiversity is the basis of this balance with implications for the farmer’s activity and the space that surrounds it, for this pollinators, vertebrates and mammals are esencial, but in a special way the insects. The destruction of the habitat of these, with the degradation of the space like hedges and other natural refuges of the insects makes them scarce, it is necessary to redefine the agricultural systems that favor the diversity in the rural space in favor to guarantee food. Agricultural farms benify sustainable activity, which is why biodiversity and agriculture must go hand in hand in order to achieve ecosystem conservation and the optimization of agricultural activity (R. Winfree and Col 2018).
The work is carried out in the Siete Lomas Protected Landscape, assessing a sustainable rural development integrating cultural heritage with flora, fauna, beekeeping, agriculture, landscape. Old and popular documentary records highlight the existence of agro-apicultural activities associated with farms, with other agroforestry activities of the time. The revitalization of the agro-beekeeping conservation activity constitutes an instrument for protecting the environment in a Sustainable Rural Development. Western Institutions have become aware of this aspect, designing programs that promote economic activities committed to biodiversity and direct business incentives that helps the conservation of the environment. In this sense the fauna and, in a special way, the bees interact with the floral mass. Its prolonged production in time favors the natural environment. Conservation agriculture and agro-ecology allow the recovery of entrepreneurial activities of vegetable production that provide food security, nourish the insects of the place contributing sustainable socio-economic values and generating new businesses that rebuild abandoned farms.

[P.21.551] Honey bee performance under different land use and conditions

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Location of bee hives play a very important role in the success of honey bee business and for profit maximization. This study was carried

out in different locations in three south western states of Nigeria: Osun, Ondo and Oyo. The sites are characterized as hill tops, residential areas with limited vegetation, farmland and thick forest. Fifteen bee hives approximately 50 meters apart were randomly placed at each location. The hives were monitored during the study period which covered both wet (May-September) and dry (November-March) seasons. Total harvest of colonies was conducted for all the hives at the end of the study period.
The results showed that hives located on hill tops and residential areas had the highest colonization efficiency of over 60% while forest areas and farmlands were both poor in hives colonization (less than 20%). Average wax production was significantly higher for hill tops and residential areas (11.65 ± 0.14, 11.75 ± 1.5 and 10.63 ± 2.0) as well as average honey (7.82 ± 0.09, 6.97 ± 0.93 and 5.78 ± 1.09) at $P < 0.05$. Honey wax and honey showed high correlation with bee hives colonization and were recorded to be highest from bee hives with highest percentage of colonization ($r = 0.89$). A number of pest discovered at the bee hives sites determined to some extent the performance of the bee hives and colonies. Occurrence of soldier ants and small hive beetle correlated positively with reduced colonization and poor yield.
In order to derive maximum profit from honey bees, adequate attention must be given to hives location as they must be located in areas with good flora supply; and free of bee pests most especially, soldier ants and small hive beetle.

[P.21.552] Bioapiculture and the joung farmers in rural development of sustainable ecotourism, Tenerife island

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The Ecological Economy related to Rural and Urban Spaces is an attraction for young farmers with a new business vision based on the bioeconomy. A current way of valuing the rural world where agroecology acquires horizons of service in agrarian farms in the form of Ecotourism defined as responsible travel to natural areas that conserve the environment, sustains the well-being of the local people, and involves interpretation and education (The International Ecotourism Society, TIES, 2015).
Acting by interacting the history of a territory with an agricultural activity is the object of a research project located in the Canary Islands, Ultraperipheral Region of Europe, of the Kingdom of Spain, in one of its Protected Natural Spaces and constitutes an ambitious challenge that has as vision generate an agrarian company integrated into the economy of the XXI Century.
Siete Lomas preserves remains of a traditional agricultural activity and has been protected by the Government of the Canary Islands; It is located southwest of the Tenerife Island, in the municipality of Candelaria, north of the Gu{imar Valley, at 1200 meters above sea level, it houses the old Chivisaya Wineries of the 19th century, abandoned and damaged by pastoral activity. A Bioeconomic resource with a history to recover where beekeeping, viticulture and forest coexisted for decades (M.L.Fuertes and F.M.Hodgson, 2018).
The history, the biodiversity and the remains of a prosperous economy of the place base the project of recovery of the space of the Bodegas. The ecological beekeeping together with the agroecology allows to favor the development of the ecosystems that surround it and constitutes the ecological soul of this integrated project in the base of the Sustainable Rural Development in the Island of Tenerife where the apieco-tourism is an alternative for the integral development of the protected agricultural space.
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[P.21.553] Keeping Stingless Bees in Costa Rica: Perspectives and Limitations

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In Costa Rica 58 species of stingless bees (Apidae: Meliponini) are known. Stingless bees are kept in wooden log hives or small boxes, with a design that is specific for each species. The stingless bee kept throughout Central America, *Melipona beecheii*, has a Maya culture related way of domestication in log hives or tree trunks, from which honey is harvested during the dry season by opening one of the extremes and the manually squeezing of the egg-shaped honey pots, collecting the honey in some recipient. Several other species are kept in boxes, being the most common *Tetragonisca angustula*, followed by *Scaptotrigona pectoralis* and Nanno*Trigona* perilampoides.
As major limitations the following factors should be considered: (1) change of land use has caused a diminishing availability of floral resources and trees with cavities where stingless bees can nest, (2) the use of pesticides on crops visited by stingless bees and for control of mosquitoes near the houses has killed many colonies of stingless bees, (3) climate change has altered the flowering period of many trees and plants and caused prolonged periods of drought diminishing the production of honey and negatively influencing the survival of colonies of stingless bees, and (4) the loss of interest in traditional and cultural values of keeping stingless bees by generations of young people.
In recent years the declining interest has been reverted and over 300 people keep stingless bees, with an estimated 1170 hives, in Costa Rica. This increase can be attributed to the development of a new and easy to apply trapping technique luring swarms of stingless bees in boxes baited with a propolis extract, which has resulted in a cheap way to obtain new hives, and because of the high value that is attributed to the honey of stingless bees, because of recent discoveries of its medicinal properties, which has raised the interest of the pharmaceutical industry on one hand and a growing gourmet and health food market on the other hand. Prices for stingless bee honey in Costa Rica oscillate between 40-80 USD per kg.

[P.21.554] Application of the Flow Hive to beekeeping with native Japanese honey bee (*Apis cerana japonica*)

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In recent years the growing interest in biodiversity and native pollinators has led to an increase in keeping native Japanese honey bees (*Apis cerana* japonica). Japanese honey bees are generally kept in traditional Japanese beehives, also known as pile box hives. These hives consist of stacked boxes that contain no frames, allowing the bees to build natural comb freely. An entire box of comb must be cut away for honey extraction by crush and strain method. Japanese honey bees have a high tendency to abscond and this disturbance increases the chances of absconsion. Additionally, the need to remove an entire box for honey extraction limits the amount of honey that can be extracted. This brings into question which hive is best suited for the native Japanese honey bee. The Flow Hive has a unique honey extraction method that allows for easy extraction with minimal disturbance to the bees. The aim of this study was to investigate the impact of the gentler Flow Hive extraction method on both rates of absconsion and honey yield. Field research was conducted comparing 35 traditional Japanese beehives to 35 Flow Hives modified for Japanese honey bees. The research was carried out by volunteer beekeepers throughout Japan. Beekeepers whose bees successfully placed honey in the Flow Frames of their Flow Hives participated in a survey. Preliminary findings regarding hive design, absconsion rate and honey extraction volume were established. There is some indication that hive design influences absconsion rate and honey yield. The most successful model was a traditional Japanese beehive with a modified top hive box containing four half-sized Flow Frames.

[P.21.555] Preservation of the Saharan bee in Algeria

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The Saharan bee (*Apis mellifera* sahariensis) would be a vestige of the time when the Sahara was fertile. It exists only in part of the Saharan Atlas which borders Morocco and Algeria. In Algeria, its range is in the mountains of AïnSefra, Bechar and BeniOunif. It is easily recognizable by its yellow color and is characterized by its softness compared to the Tellian bee (*Apis Mellifera* intermissa).
If Algerian beekeepers know very little about this bee, it is because it has become very rare.
Why? Several factors seriously hinder its development, which are in order of importance:
- hybridization with an invasive species such as the Tellian bee;
- the introduction of diseases and parasites introduced by the Tellian bee;
- predators such as the African Bee-eater;
- poisoning due to locust control;
- lack of honey resources due to drought;
- the massacre of colonies by honey hunters.

The two most urgent steps to be taken in parallel to save the Saharan bee are to multiply it massively while prohibiting the introduction of the Tellian bee into the natural cradle of the Saharan, the region of Ain Sefra. It's the conservation in situ.
It is in this context that the Division of Research in Animal productions of the INRAA and the University M'Hamed Bougara collaborated to launch a research project underway for rehabilitation outside the natural craddle of the Saharan Bee (ex-situ). The region of Adrar where the risk of genetic pollution is minimal is well suited for that. The Adrar station of INRAA will play the role of fertilization station of the Saharan bee.
The originality of the project is to rehabilitate a race of bees facing the threat of extinction. This rehabilitation program of the Saharan bee will first of all preserve an exceptional Saharan fauna heritage that has taken thousands of years to adapt to its hostile environment. To reach that objective, it would be necessary for the region of Aïn Sefra to be constituted conservatory of the Saharan bee.

[P.21.556] Current conservation status of *Apis mellifera caucasica* and *Apis mellifera anatoliaca* apiaries in Turkey

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Honey bees are important pollinators of agricultural activities, providing a keystone ecosystem services and recent massive colony losses made honeybees one of the most important insect species to take measures for their survival. And many scientists from all over the world raised the awareness for the conservation of this unique species for the future of our world. Thus many countries are now implementing conservation strategies to preserve their indigenous honeybee subspecies. Two subspecies namely *Apis mellifera anatoliaca* and A. m. caucasica and several ecotypes of these subspecies are under protection in conservation apiaries in different parts of Turkey. By the years, these conserved apiaries are questioned whether they are still keeping their original status? Also due to threats A. m. caucasica in Camili-Artvin is used to establish a replica conservation apiary outside the original conservation site, and thus the second question also raised whether this replica conservation apiary represents the original population or not after years? In this work we sampled honey bee colonies from Ankara (A. m. anatoliaca), Camili, Hatila, and Posof (A. m. caucasica) for morphometric analysis after five years, in order to answer the

questions rose previously. A total of 26 characters belonging to fore wings, hind wings, legs, and color of tergites were used in multivariate statistical analysis. Along with standard morphometrics, we also applied Geometric Morphometric approach using 20 landmarks on the forewing to answer the questions. The results of both analysis showed that the original populations from conservation apiaries were keeping their original measurements and also the newly established (a few years ago) replica conservation apiary of Hatila-Artvin was overlapped with the original conservation site (Camili-Artvin) providing that the newly established replica conservation apiary represents the original population. Future considerations and projects are also discussed for the conservation efforts on Anatolian and Caucasian honeybees in Turkey.

[P.21.557] Sustainable Rural Development and Biodiversity Conservation with Bees (Apis mellifera & native stingless bees)

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In Madre de Dios, CEDE implements a beekeeping model with Apis mellifera and native stingless bees for the sustainable use of the Amazon rainforest, contributing to i) sustainable rural development improving the living conditions of rural and marginal-urban population and indigenous peoples, ii) food security and iii) biodiversity conservation.
Beekeeping is an excellent alternative for family enterprises management and social inclusion programs.
Management and breeding of Apis requires more care, dedication, time, and inputs than stingless bees. However, Apis has more adaptation abilities to different ecological levels and climates, making viable its breeding in more extreme and varied environments (e.g.: Andean highlands). The most favorable environment for stingless bees is the tropical and subtropical region of the American continent.
Both species generate goods and services, contributing to improve the living conditions of rural and marginal-urban population. Bee products, such as honey, pollen, propolis, wax, and other byproducts, are of great economic importance. In Peru, in the fields dedicated to export agriculture, beekeepers install and rent Transhumant Apiaries, providing pollination services to increase agricultural productivity and improve the quality of products in size, uniformity, color/appearance, amount of micronutrients.
In Madre de Dios, honey is important for human health and food security of families as part of the struggle against anemia. Apis mellifera produce more honey and other products; however, the nutritional and medicinal value of honey and propolis of stingless bees is significantly higher.
Thus, CEDE includes the study of the adaptation abilities of stingless bees through the installation of Apiaries in homes, schools, and public places, which, due to their docility, could be handled easily, including by children, without discomfort or risk of stings. In this way, children will have access to honey as part of their dialy diet.
In the Amazon, biodiversity conservation is based on the pollination carried out by stingless bees and other insects, promoting greater production of fruits and seeds, and ensuring the proper balance of the forest.

[P.21.558] Scientific Beekeeping a Gold Mine for Sustainable Agriculture and Rural Development with Special Reference to India

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Beekeeping is an agro-based activity which is being undertaken by farmers/landless labourers' in rural area as a part of Integrated Farming System (IFS) and promoting sustainable and eco friendly agriculture. Honeybees are best known for the honey they produce, their economic role in nature is to pollinate millions of flowering plants and ensure setting of seed/fruit and offering services to the society through ensured pollination as well as by providing honey and a variety of beehive products Beekeeping also helps in sustaining bio-diversity and environment sustainability. Beekeeping is one of the best income & employment generating activity for rural development and serves as a source of livelihood for rural poors. Scientific beekeeping may help in minimizing Human and Elephant conflicts.
2. *Apis mellifera*, *Apis cerana*, *Apis indica*, *Apis florea* & stingless bees are major bees found in India. Indian beekeeping is based on *Apis mellifera* & *Apis cerana*. India is one of the main honey producing & exporting countries in the world. The diversified agro climatic conditions and floral availability in India provide great potential and opportunities for beekeeping. To pollinate 12 major agricultural / horticultural, India need 200 million honey bee colonies.
3. The Ministry of Agriculture and Farmers' Welfare, Govt. of India is promoting scientific beekeeping in big way and National Bee Board (NBB), is the Nodal Agency for promotion and development of scientific and robust beekeeping in the country. NBB has been involved in implementation of several projects / activities including, HRD Programmes, publications, road map for scientific beekeeping, developing training modules, preparing & circulating Advisories on Good Beekeeping Practices (GBPs), identifying issues / agenda for beekeepers' driven research, developed system for traceability source of honey and other beehive products / registration of beekeeping industry, implemented PPP Project with QSI, Breman, Germany, quality nucleus stock, setting up of Integrated Beekeeping Development Centres (IBDCs), bee disease diagnostic & products quality control labs. Recently, Indian Govt. notified strong standards for honey, bees wax & royal jelly. India can offer best quality honey and other beehive products as well as organic beehive products to the world.

[P.21.559] Conservation of Mexican Biodiversity through stingless bees as pollinator agents in indigenous Totonac communities

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In the latest news there is a global concern about the state of bees (Allen et al., 1998; Kearns et al., 1998) due to a growing devastation of natural habitats. Which increases the concern of specialists to observe the direct impact on populations of native bees that have not been domesticated, mostly, solitary habits that build nests in floors, walls and / or trunks (Nates, 2000; Roubik 1983, 2006; Nogueira-Neto 1997; Eltz et al. 2003; Oliveira, 2012). Stingless bees have been known for their production of honey, propolis and pollen. Today its role as providers of ecosystem services is recognized (Roubik 1989; Michener 2007; Freitas et al. 2009). Today more than 600 species in 56 named genera live in tropical and subtropical areas of the world. There are 400 known species in the Neotropical region and it is estimated that there are more to be described (Cortopassi et al., 2006, Roubik 1983). The stingless bee *Scaptotrigona* mexicana is widely distributed throughout the Mexican tropics and includes economically important species used in meliponiculture. The case of *Scaptotrigona* mexicana, is one of the stingless bees traditionally used for the production of honey in Mesoamerica (Albores- González et al. 2011). The importance of the investigation recovers the worldview that frames the system of analysis of the ecological environment and the ways of relating to it by the Totonac indigenous communities. The above intervenes in their productive practices and their forms of interaction with biocultural resources. Allowing to establish the potential combinations of practices derived from traditional ecological knowledge with those derived from scientific knowledge in a way that can result in sustainable development modalities. The research consists of analyzing the cognitive system of the communities, the repertoire of their ecological knowledge and sustainable practices where the stingless bee is located, symbol of the Totonacapan known as taxkat (t'áxkat), a bee that lives inside the trees, on land and in rocky cavities.

[P.21.560] University of Minnesota-Nicaragua Bee Exchange

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The University of Minnesota Bee Lab-Nicaragua Bee Exchange program was established to provide opportunities for beekeepers and researchers to learn about beekeeping outside of their geographical context. This presentation will share the University of Minnesota's experience building cross-cultural relationships among beekeepers as well as highlight differences in beekeeping due to climate, flora, genetics, culture, and economics. By showcasing this successful cross-cultural exchange, we hope to inspire others to pursue similar initiatives that will bring benefits to bees and beekeepers around the globe.
The University of Minnesota Bee Lab conducted two trips to Nicaragua to share beekeeping experiences and foster opportunities for beekeepers to learn from each other. The goal of the first visit was to establish relationships with beekeepers in Nicaragua, participate in meaningful dialogue, and identify opportunities for future exchanges. In January 2017, 8 employees from the University of Minnesota Bee Lab traveled to Nicaragua to engage with beekeepers. During this inaugural visit, many beekeepers in Nicaragua expressed interest in participating in beekeeping workshops. In March 2018, 10 bee scientists, beekeepers, and bee enthusiasts from Minnesota and Michigan traveled to Nicaragua to conduct a 2-day beekeeping workshop, continue to build relationships with beekeepers in Nicaragua, and facilitate opportunities for everyone involved to learn about bees.
Although the beekeepers from the United States were not experts in managing Africanized honey bees or honey production in Nicaragua, their collective professional knowledge and experience demonstrated that providing education about honey bee biology to beginning beekeepers can lead to improved beekeeping skills. Wild bee experts in the group connected with stingless beekeepers and environmentalists to identify local bee species and discern opportunities for future research projects. Partners in Nicaragua taught the group about Africanized honey bees and their management, challenges and opportunities of beekeeping in Nicaragua, and beekeeping with stingless bees.
Hopes for future exchanges include bringing Nicaraguan beekeepers to Minnesota or Michigan so they may also get to see beekeeping in a different geographical setting. The University of Minnesota and Michigan State University continue to engage with beekeepers in Nicaragua through virtual beekeeping exchanges.

[P.21.561] Beekeeping with local and indigenous bees

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The quality of genetic material is one of the success factors of market agriculture. Beekeeping with local or indigenous breeds is nowadays only found in the low industrialized countries where the notion of business is not yet entered into mores. With local breeds what are the strengths and weaknesses of beekeeping? Among its strengths one can mention the protection of honey bee which is more and more subtracted from the misdeeds of picking; this situation reinforces the contribution of this main pollinator to the conservation of biodiversity and to the increased agricultural yield of these environment.
Among its weaknesses one can indicate the low knowledge of its performance because little seriously studied due to its great variability or the ignorance of its interests. This may explain the quite total lack of professional beekeeping in these under developed countries.

If the use of local breeds could yield as obtained in our station a performance of 30 kg of honey per beehive per year, it could be said that the low productivity often spoken for native beekeeping is not only the fact of race but also and above all related to the problems of the beekeeper' skill and the quality of the bees' grazing. Beekeeping with local and indigenous breeds can still play a major role in the take-off of the economies of under developed countries if one tries to well understand it and better maintain it. It also appears as a guardrail in the face of the international trend of selection, particularly vis-à-vis of the production of GMOs whose threats to human health and the disappearance of living species have been taken very seriously since a while.

[P.21.562] Exploring the native and imported bee species of Bhutan by Paula Carnell

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On a solo visit to Bhutan in November 2018, Paula Carnell met up with the Beekeeping Cooperative in Bumthang, Bhutan to understand more of the agricultural departments policies on honey production and beekeeping in relation to native and imported bee species.
She learned that 5 colonies of *Apis Mellifera* were imported in 1998 and have now expanded to 1000 colonies spread across central Bhutan managed by local beekeepers.
In the south, *Apis cerana* bees are still kept in log hives and hives built into the walls of honey houses. During a visit to the south of Bhutan, near to Tserang, Paula met with such a beekeeper. Paula also travelled from Paro to Phunacka across the country finding *Apis dorsata* and Laboriosa in colonies attached to cliff faces and the window frames of temples and monasteries. These were untouched by honey harvesters.
The beekeeping Cooperative is encouraging the production of honey from their growing number of *Apis Mellifera* beekeepers but with limited domestic market due to the Buddist belief of taking and eating honey being a sin.
Paula discusses the as yet unresearched impact of the introduced species on solitary and bumble bees in Bhutan, the success of the disease resistant *Apis Mellifera* and the countries organic farming status in relation to bee health. The culture of traditional medicine also being available through the Kingdom's hospitals is also related to the overall health and wellbeing of the kingdom's nature and human population.
Paula plans to return to Bhutan with a research team and film crew to understand more of the beekeeping and bees of Bhutan, along with taking lessons from Bhutan to share with the beekeeping community globally.

[P.21.563] Evaluation of *Melipona seminigra* colonies in the Carajas Nacional Forest: seasonality of brood combs, honey, and pollen pots

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The bee Melipona seminigra plays an important role in seeds and fruits generation in the Amazon forest. Traditionally, this bee species is also important for local populations' income through honey production. From January to December 2018, we evaluate colonies of M. seminigra installed in the Carajas Nacional Forest in three locations with different degrees of anthropization: (A1) inside a pristine forest; (A2) close to human facilities (buildings and roads), but surrounded by pristine forest; and (A3) a forest bordering a large deforested area. We measured the size of the brood area and we counted the number of honey, and pollen pots present in the colonies. Data were analyzed using Kruskal-Walis + Tukey posthoc and Wilcoxon tests. At all locations tested, two colonies got weak and one got strong (KW, $p< 00.5$). Comparing the strong colonies from each place, we found significant differences in the size of brood combs (KW, $p = 0.04$), larger in A2 than in A3 (Tukey, $p = 0.03$). The number of pollen and honey pots was higher in A2 than in all others (Tukey, $p < 0.01$) and it was similar between A1 and A3 (Tukey, $p > 0.58$). Moisture, competition, and parasites could be involved in the bad performance of the colonies installed in A1. In all three localities, the brood area increased from July until October/November, when it decreased again. The honey storage peaked in August and pollen storage increased in October and November.

[P.21.564] Beekeeping as One of the Important Branches for the Development of Mountainous Rural Georgia

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The mountainous regions of Georgia with rich traditions in agriculture are now almost deserted. One of the most important steps to stop this is the development of beekeeping, which can be easily achieved with mountain gray bee (*Apis mellifera caucasica*) famous for its biodiversity. It is represented by several populations in our country (Megrelian, Guruli, Kartli, Abkhazian). They are found in both Eastern and Western Georgia. It is noteworthy that Georgian populations are distinguished by a number of positive features compared with other species in the world, such as meekness, less forage consumption, maximum use of a working day, extraction of nectar from deep nectarines, etc. Despite the unstable climatic conditions in the mountainous regions, these populations produce on average 18-25 kg honey, 4-5 built combs (wax productivity). Their average daily egg production is 1200-1800 eggs. Natural and ecological conditions in the traditional area of their distribution provide the possibility to arrange biobeefarms, without much effort, on the basis of the introduction of

the modern technologies of bee keeping. Besides, in the remote mountain areas no technogenic pollutants, pesticides or genemodified plants are found.
All the above will contribute to the increase of the share of bio-products in beekeeping, followed by sustainable development of agro-ecotourism and agriculture. This will lead to the settlement of mountain residents back to their villages, more jobs and improved socio-economic conditions of low-income population.

[P.21.565] Addressing Challenges Facing a Beekeeping Development Project in Southeast Madagascar

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The Malagasy honey bee *Apis mellifera* unicolor is a subspecies of the western honey bee which is endemic to the island of Madagascar. Interestingly, unlike the neighbouring subspecies in East Africa, A. m. unicolor has not yet developed tolerance in response to the recent introduction of Varroa destructor. Problems with varroa infestations, therefore, compound existing challenges shared by beekeepers in other developing countries, including limited access to apicultural materials and high rates of deforestation. SEED Madagascar (SEED) is an award-winning UK-based NGO which has worked in the Anosy region of Southeast Madagascar for nearly 20 years. SEED's Project Renitantely (meaning honey bee in Malagasy), aims to promote beekeeping as a sustainable livelihood for 78 farmers and fishermen in rural communities. Using hands-on demonstrations and creative visual learning aids to overcome the barrier of largely illiterate audiences, Project Renitantely holds beekeeping workshops across 6 target communities. Additionally, the project helps establish secure routes to market, encourage women's participation in beekeeping, and advance understanding of the status of varroa in the region. SEED is currently developing a second phase for Project Renitantely, working with beekeepers to develop a robust, contextually appropriate strategy to combat varroa and conserve this endemic subspecies, while striving to alleviate poverty through rural development.

[P.21.566] Beekeeping and Rural Development

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The destruction of farm crops by elephants from the Kakum National Forest in the Central Region of Ghana is a big problem to the locally affected farmers.It causes problems between the park management and the farmers when they need the cooperation of the farmers within the loyalty to help in the protection of the flora and flaura hantswithin the park.The farmers need to protect their crops and have even employed hunters to kill the marauding elephants. It would be better to discourage the elephants from coming out if the forest and thereby improving the cooperation between the park management and the local communities.It has been shown that elephants will not approach beehives and this can be used as a barrier or fence between forest and the farm crops. Beekeeping will also be an income generating activity for some of the local farmers as well as helping with pollination both within and outside of the forest thus maintaining biodiversity and reducing pressure on the forest.

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