OP-088 [Bee Biology]

A dynamic picture of the diffusion of Apis mellifera mitochondrial DNA lineages in Italy over the last few decades utilizing honey as a source of honey bee population genetic information

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Animal and Food Genomics Group, Department of Agricultural and Food Sciences, University of Bologna, Bologna, Italy

The genetic integrity of the European *Apis mellifera* subspecies in many European regions has been threatened by the human-mediated dispersion of non-native populations and lines, which has intensified over the last few years. Many studies have utilized mitochondrial DNA (mtDNA) analysis from honey bees to gather information on the diffusion of various mtDNA haplotypes and to monitor the genetic integrity of *A. mellifera* subspecies. Honey contains environmental DNA (eDNA) traces from all organisms directly or indirectly involved in its production, including the DNA of the honey bees that produced it, making it useful for indirectly monitoring the genetic integrity of honey bee populations. In this study we present a snapshot of the diffusion of the main *A. mellifera* mtDNA lineages in Italy from the last few decades using honey as a source of honey bee mtDNA sequences. DNA was extracted from over 5000 honey samples produced in all regions of the Italian peninsula, Sardinia and Sicily, from 1985 to 2024. PCR products were analyzed using a fragment size-based assay. Results confirmed that the C lineage was the most frequent mtDNA haplotype throughout Italy except in Sicily where the A lineage was highly represented. Other mtDNA haplotypes (A and M lineages) were present in almost all Italian regions. The frequency of the A mtDNA lineage has recently increased in many Italian regions, indicating an increasing level of genetic admixture within the Italian honey bee population. The results provide a time-dynamic distribution map of honey bee mtDNA lineages in Italy that can be valuable for designing and evaluating the potential effectiveness of conservation policies aimed at maintaining the integrity of honey bee genetic resources in Italy. Additionally, we also demonstrated that honey offers a simplified yet extensive population genetic picture, as each honey sample may contain the DNA of many different honey bees from different colonies or even different apiaries.

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Animal and Food Genomics Group

A dynamic picture of the diffusion of *Apis mellifera* mitochondrial DNA lineages in Italy over the last few decades utilizing honey as a source of honey bee population genetic information



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COPENHAGEN

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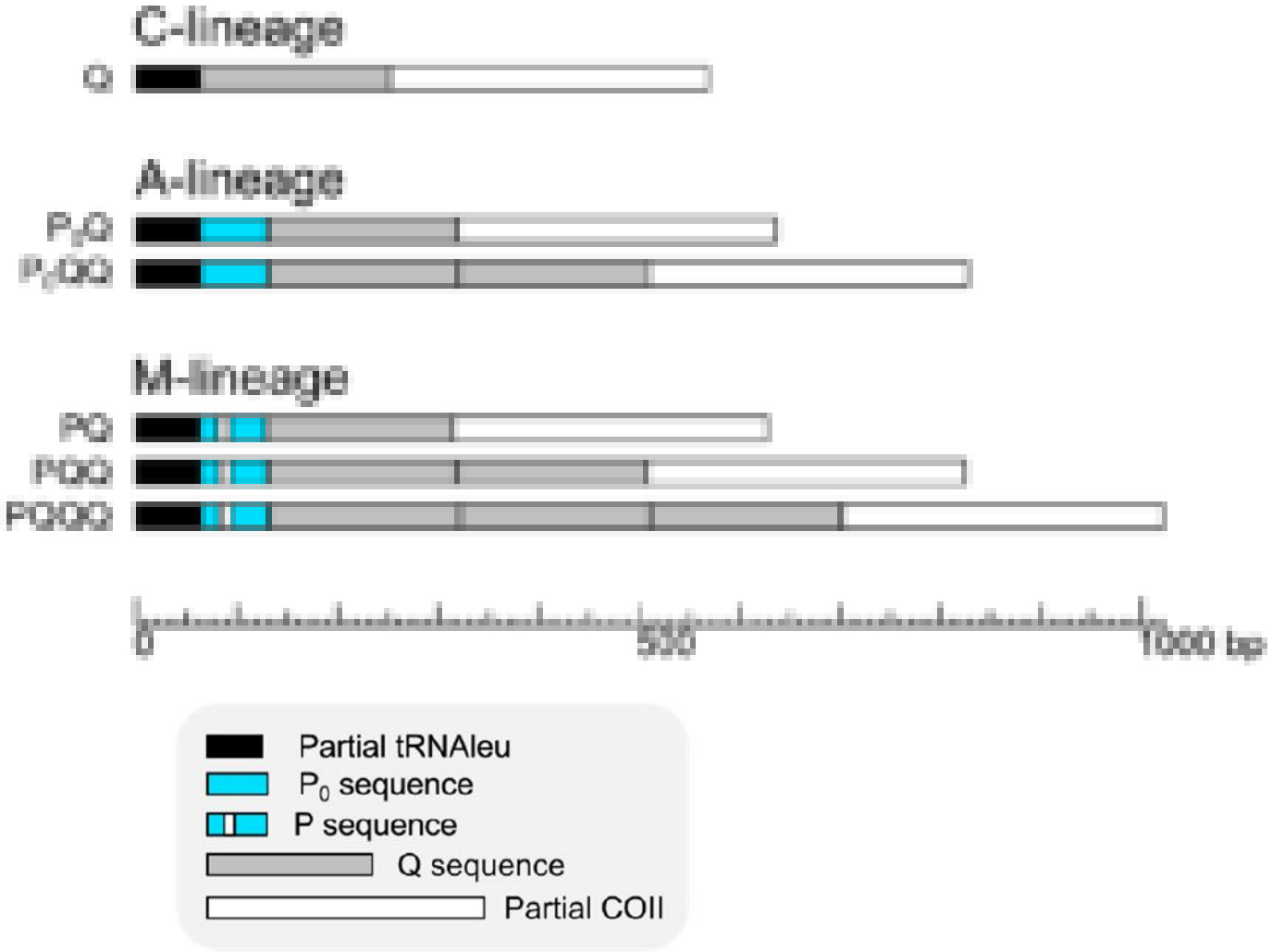
- The conservation of the genetic integrity of *Apis mellifera* subspecies is a matter of concern

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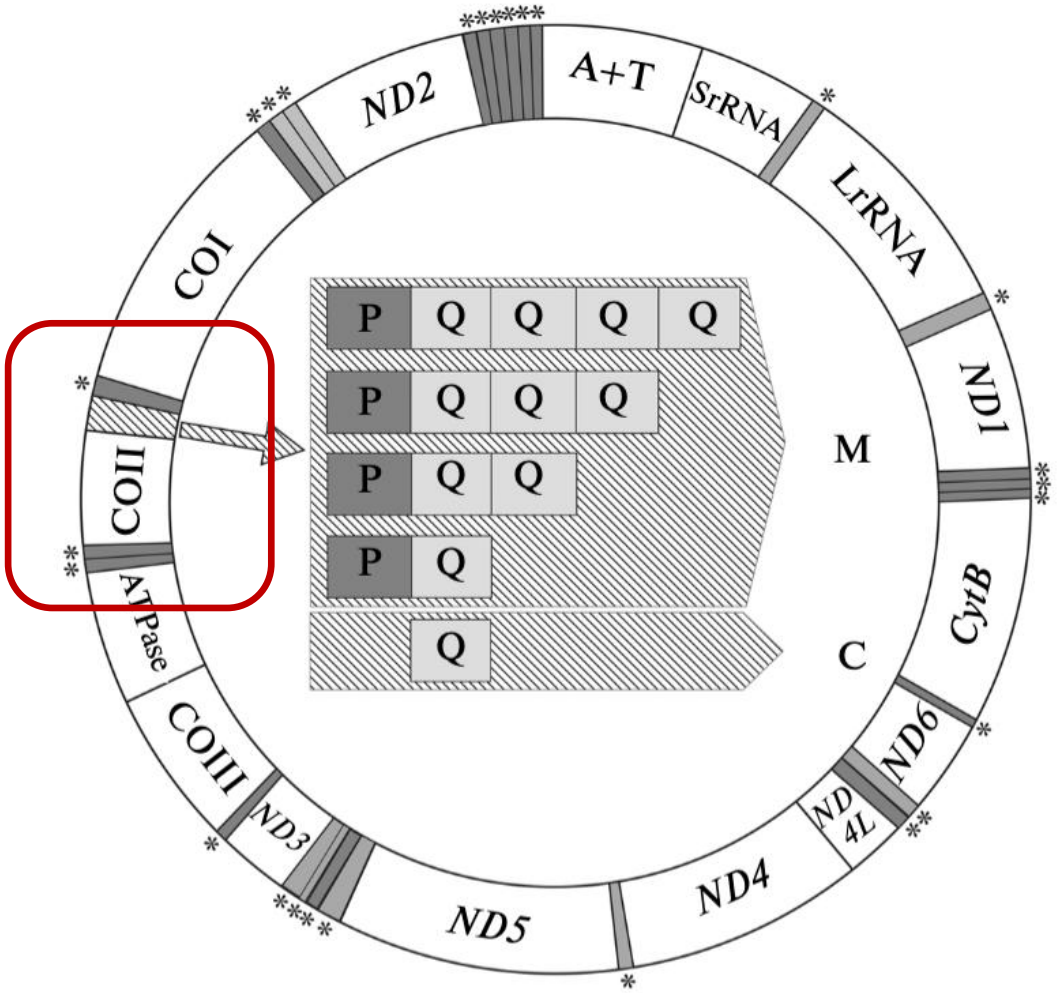
- Population genetic studies should be conducted to monitor honey bee populations

- The conservation of the genetic integrity of *Apis mellifera* subspecies is a matter of concern
- Population genetic studies should be conducted to monitor honey bee populations
- Mitochondrial DNA can provide the simplests genetic information

tRNALeu-COII mtDNA region



Oleksa et al., 2021



Ilyasov et al., 2015

Apis mellifera lineages



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Apis mellifera subspecies in Italy

Apis mellifera lineages





A. m. mellifera

M

A. m. carnica

C2



A. m. ligustica



C1 (& M7)

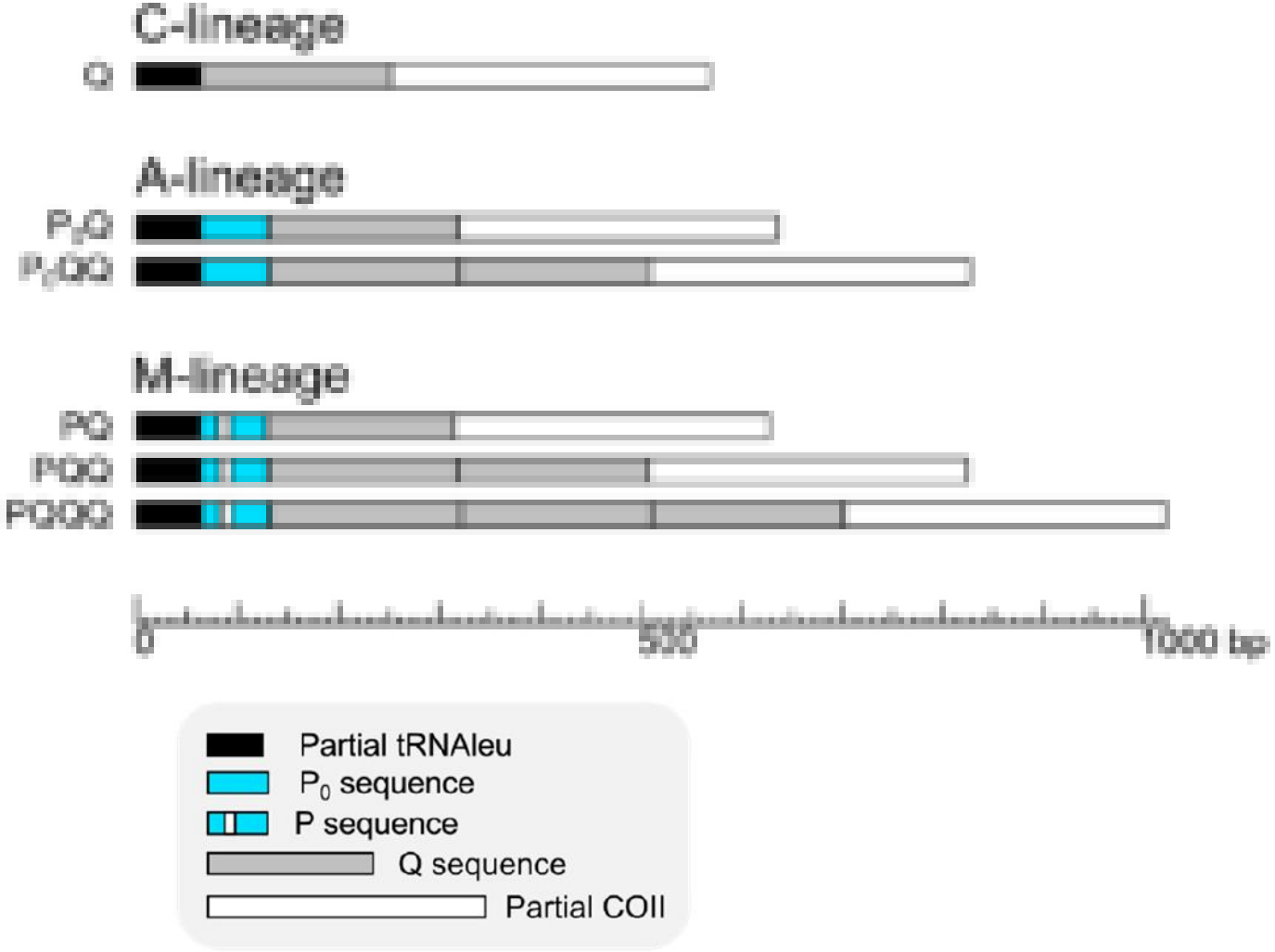


A

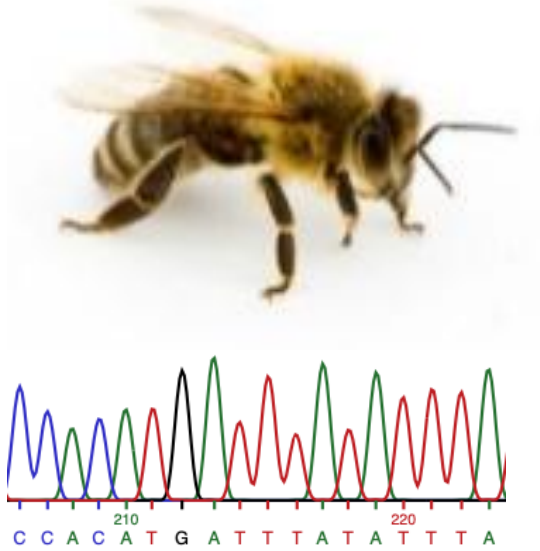
A. m. siciliana

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tRNALeu-COII mtDNA region

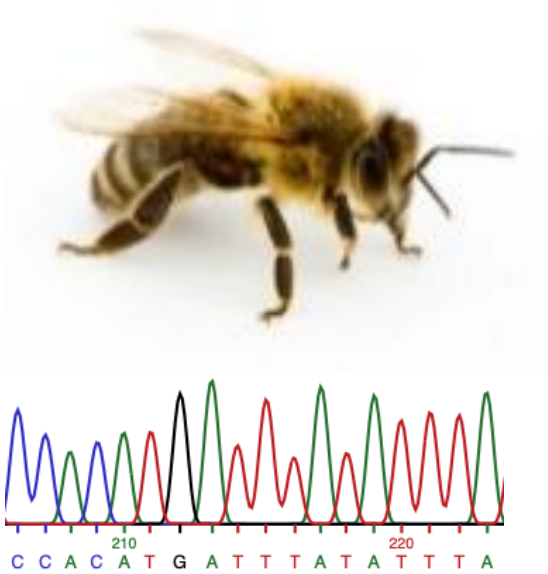


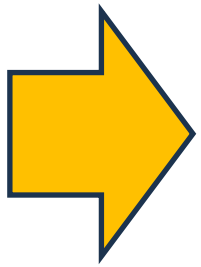
Oleksa et al., 2021

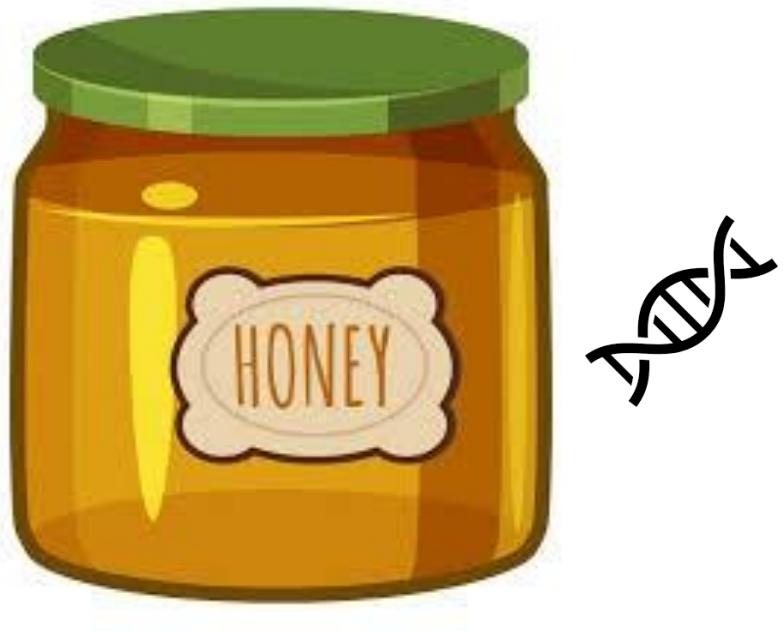


- Cost-effective large scale monitoring approaches are needed to obtain a more detailed genetic picture of *A. mellifera* populations

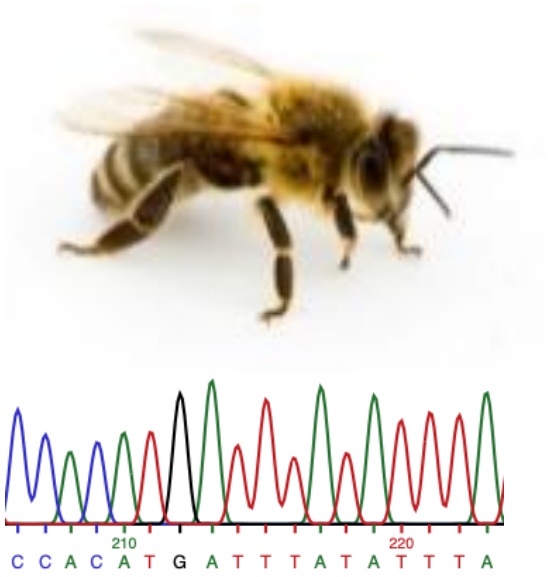
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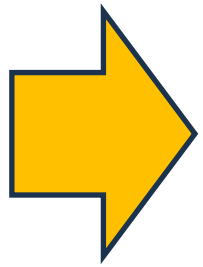




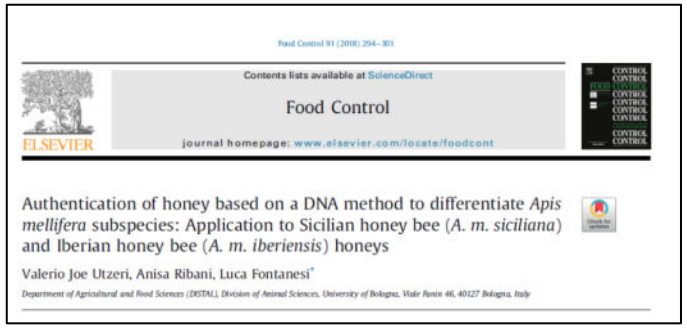


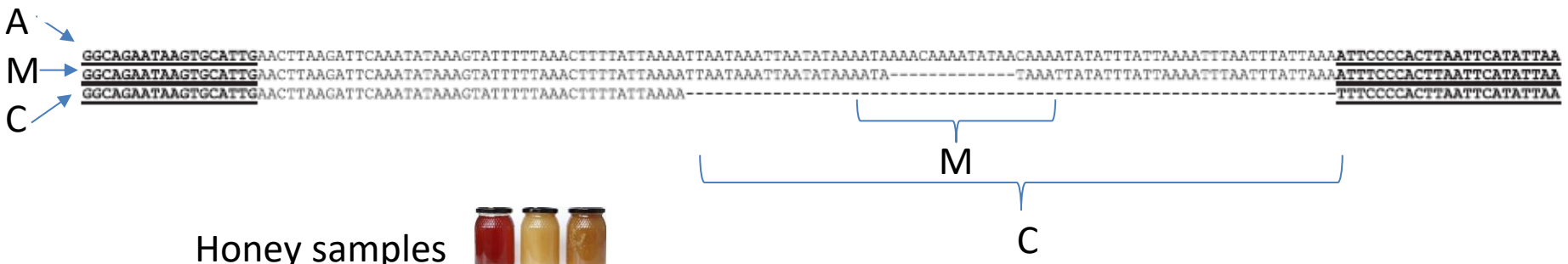
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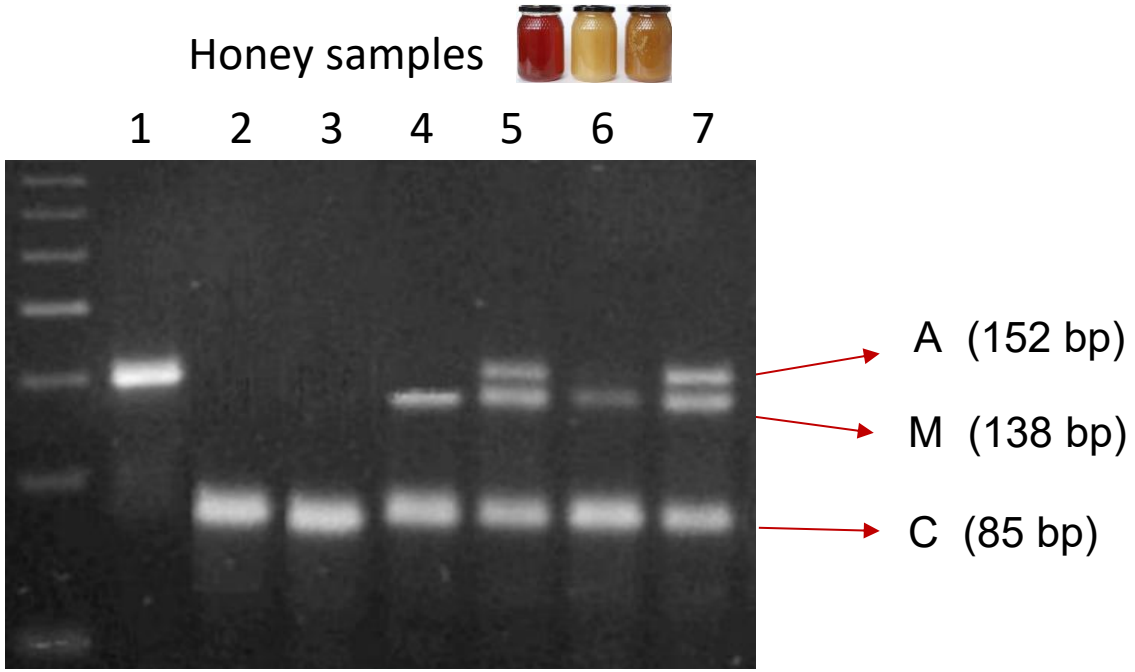


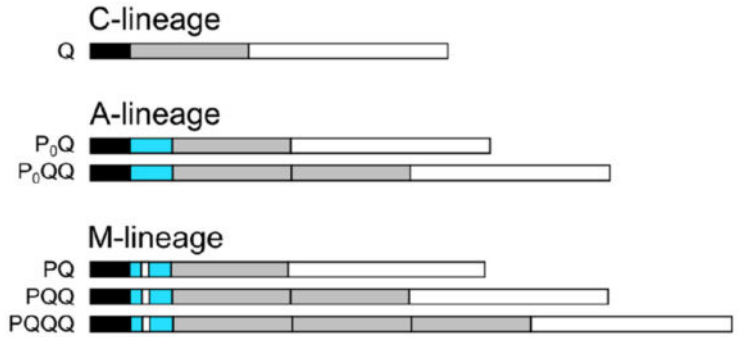






PCR + agarose gel electrophoresis





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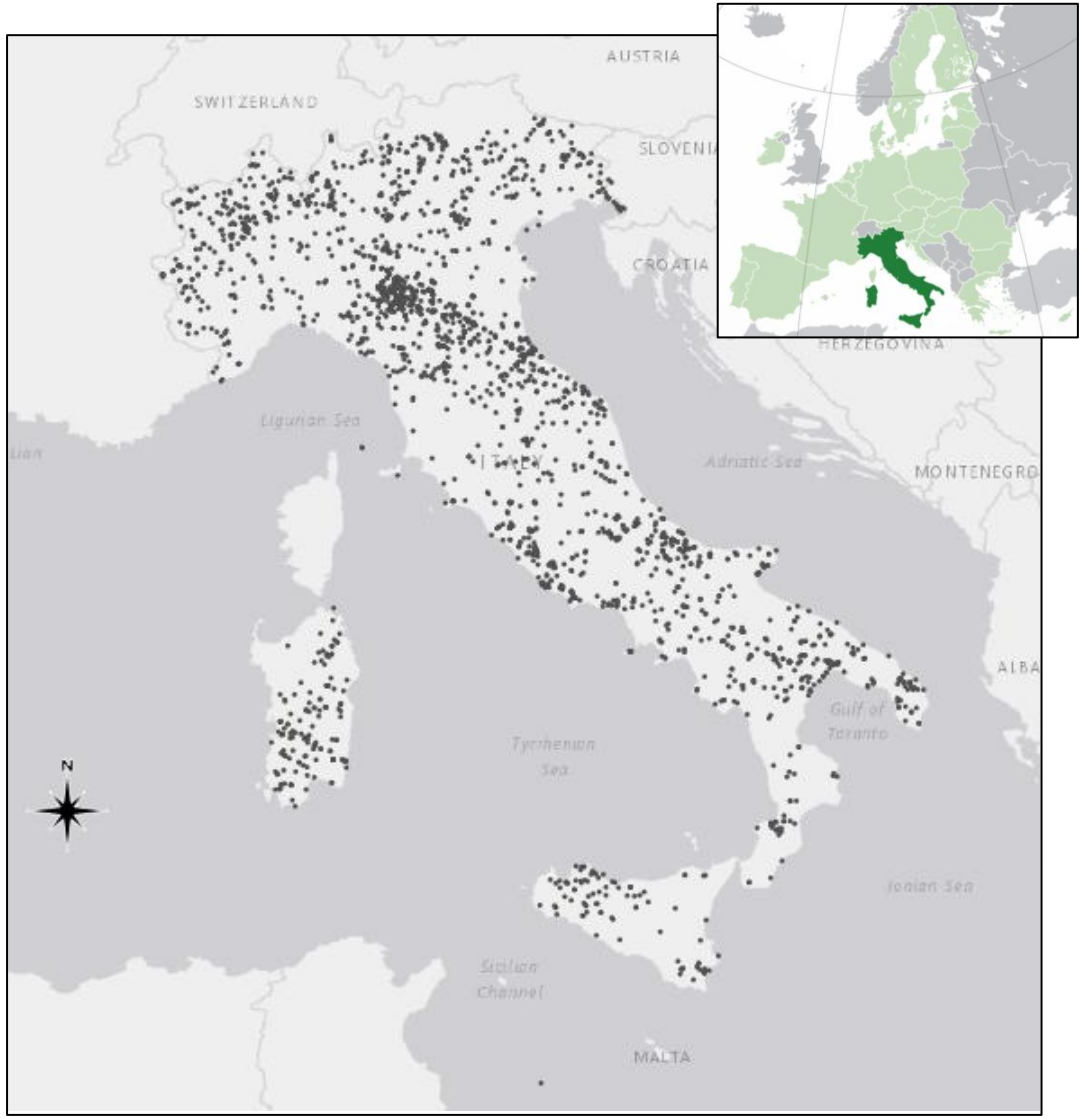
Aim

To obtain an updated an over time high-density distribution map of honey bee mtDNA lineages covering the entire Italy 🇮🇹 using honey as a source of honey bee DNA



Materials

- 4,303 honey samples
- **Produced over the last >35 years**

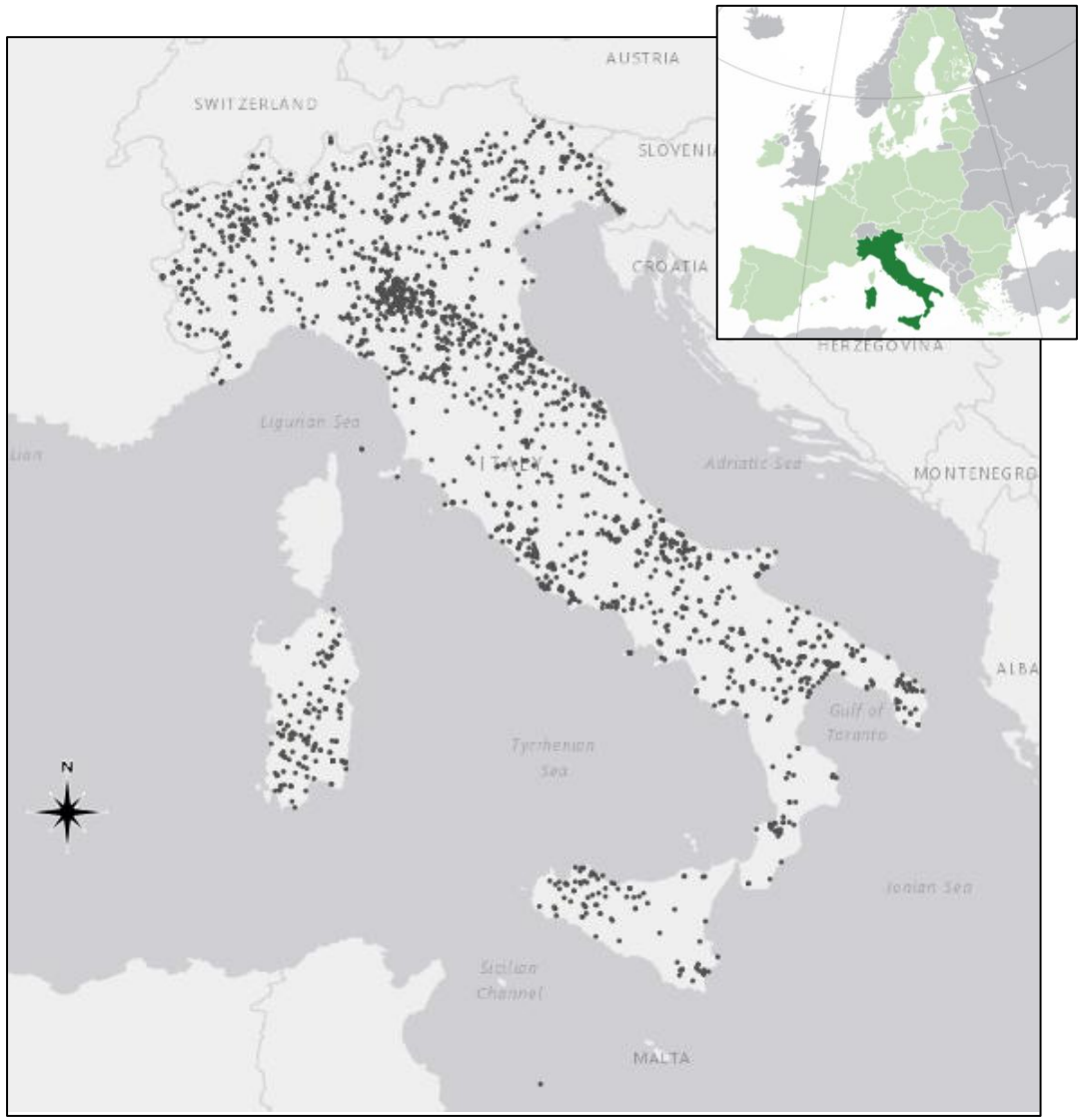




Materials

- 4,303 honey samples
- **Produced over the last >35 years**
- **Old samples (1986-2017)**
- **Recent samples (2018-2023)**

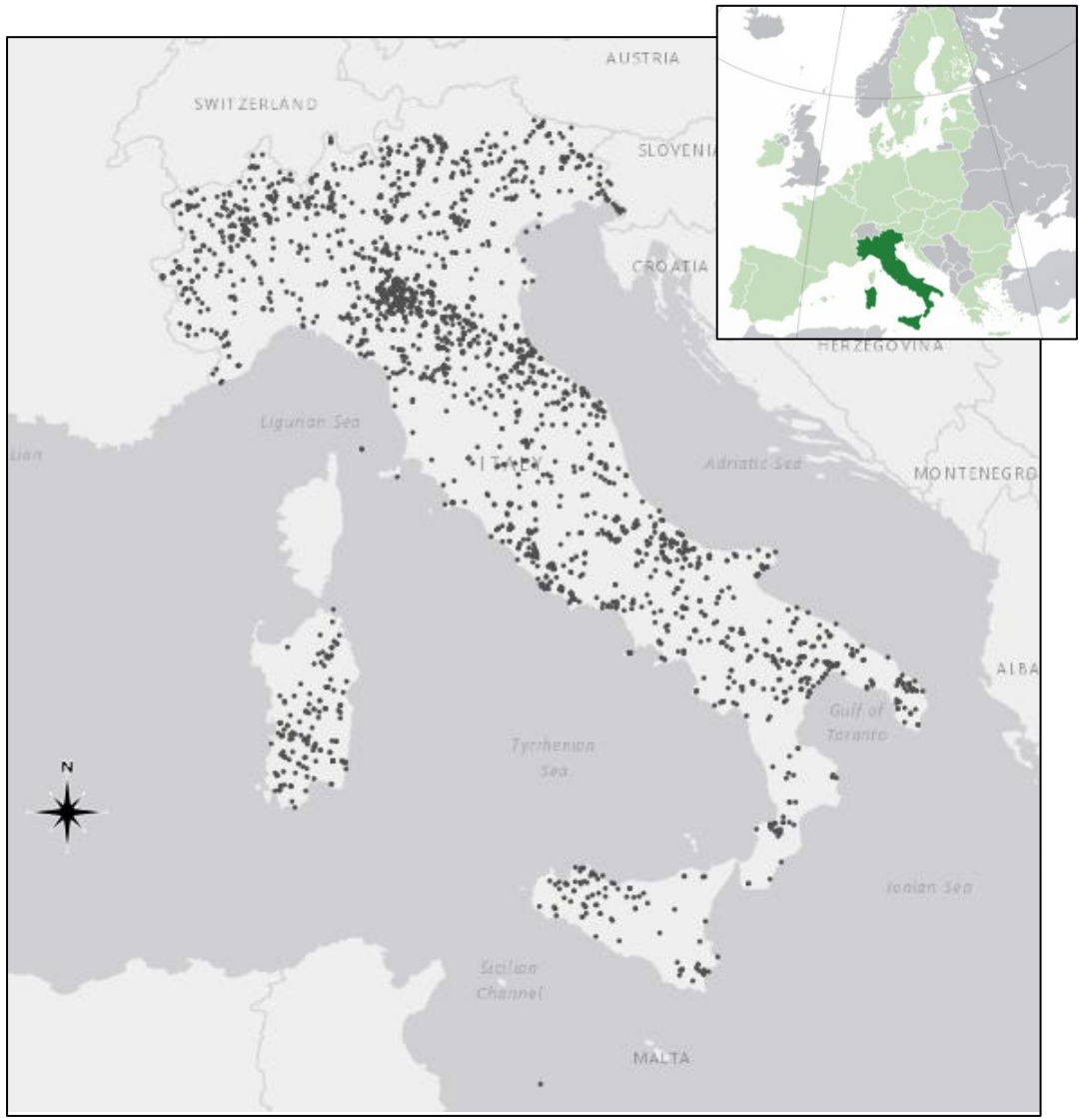




Materials

- 4,303 honey samples
- **Produced over the last >35 years**
- **Old samples (1986-2017): no. 177**
- **Recent samples (2018-2023): no. 4126**

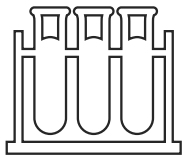


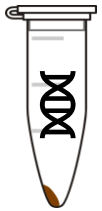


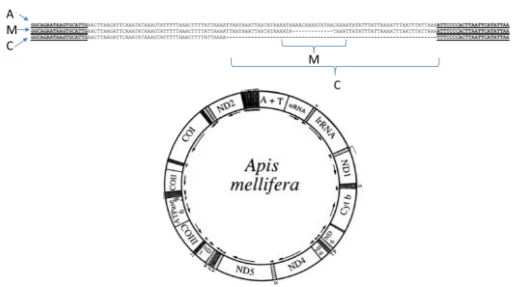
Methods

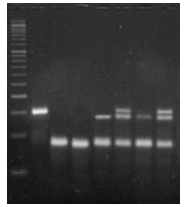


Honey pre-treatment → eDNA extraction → PCR *tRNALeu-COII* → 4% agarose gel







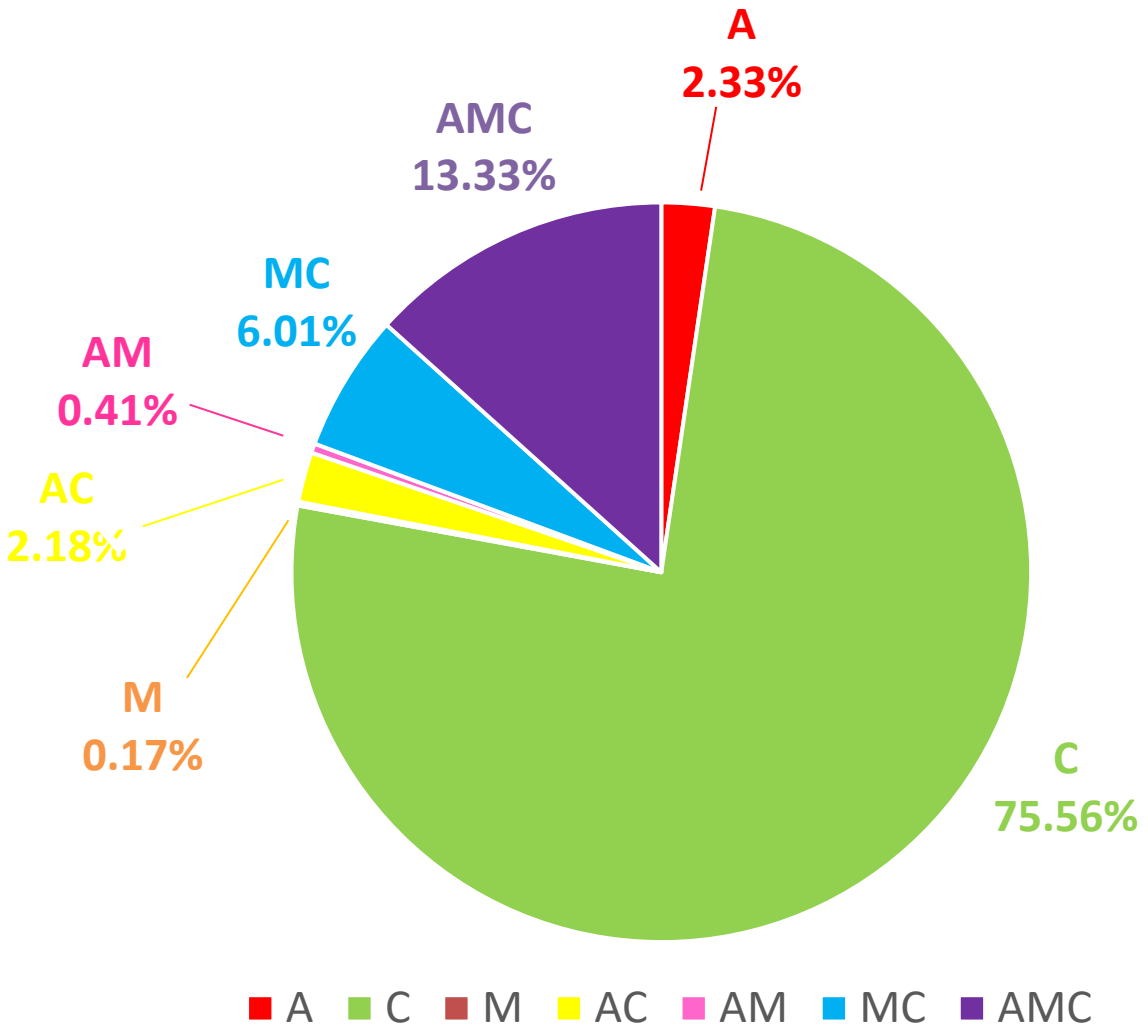


Results

2018-2023 (all 6 years)

- C
- A
- M
- AC
- AM
- MC
- AMC

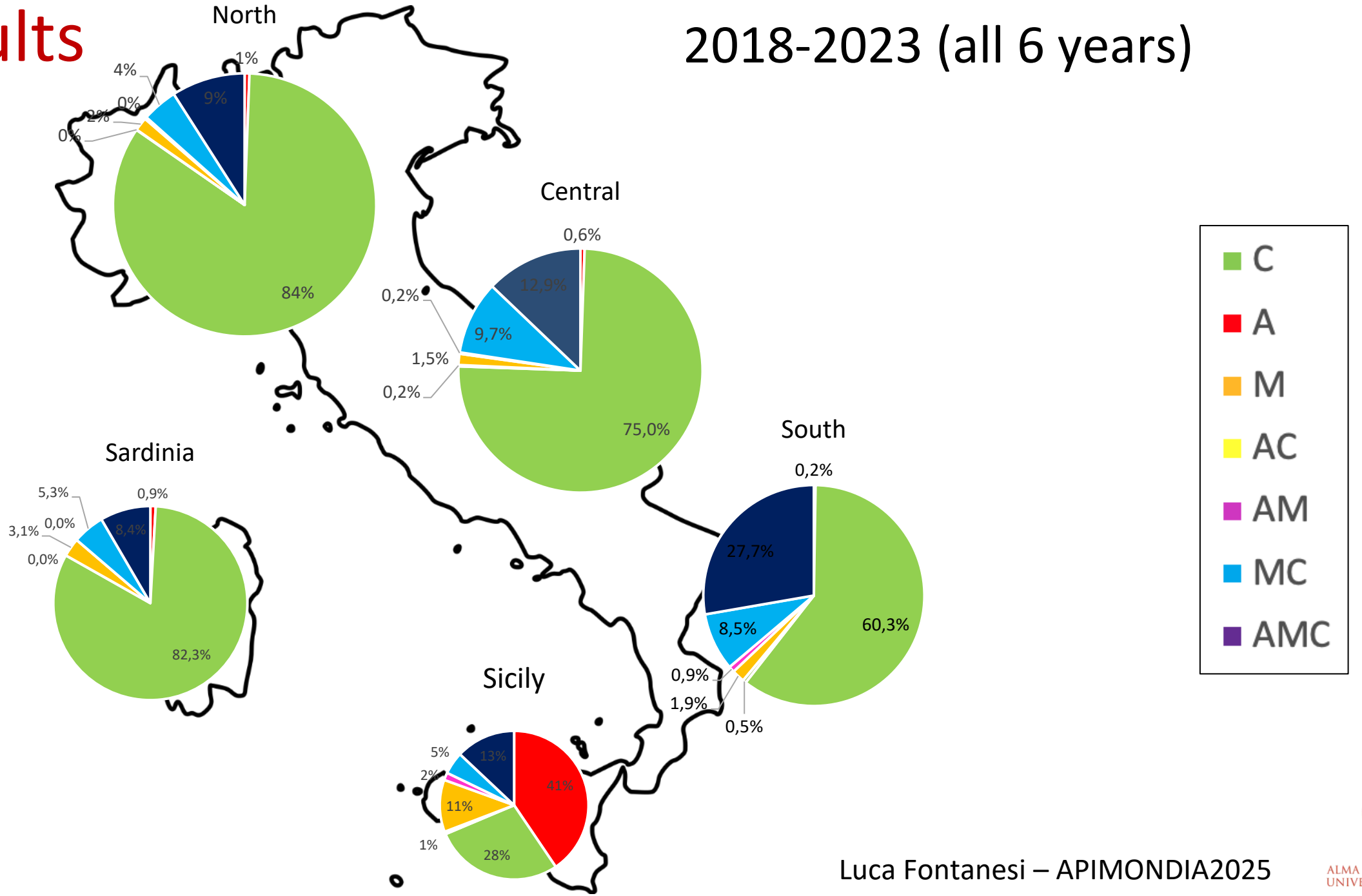




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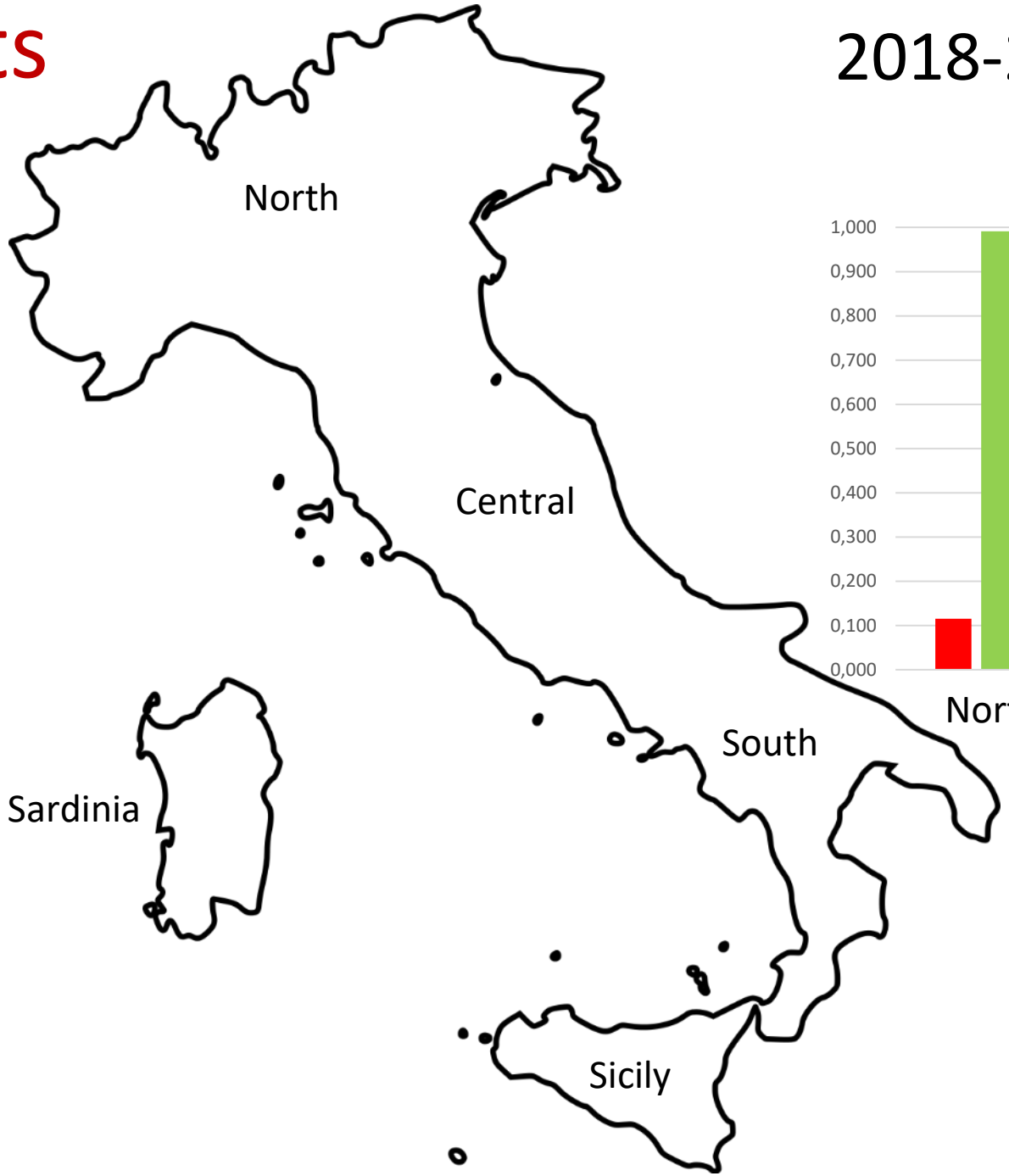
Results

2018-2023 (all 6 years)

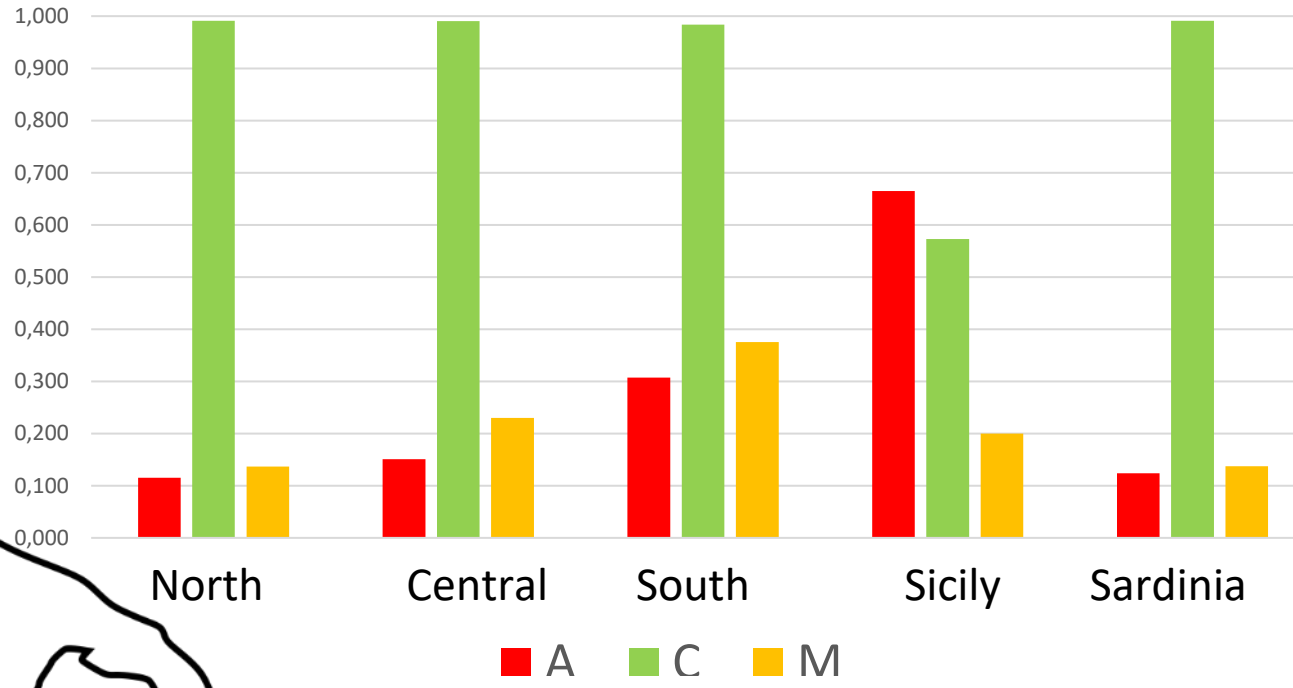


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Results



2018-2023 (all 6 years)

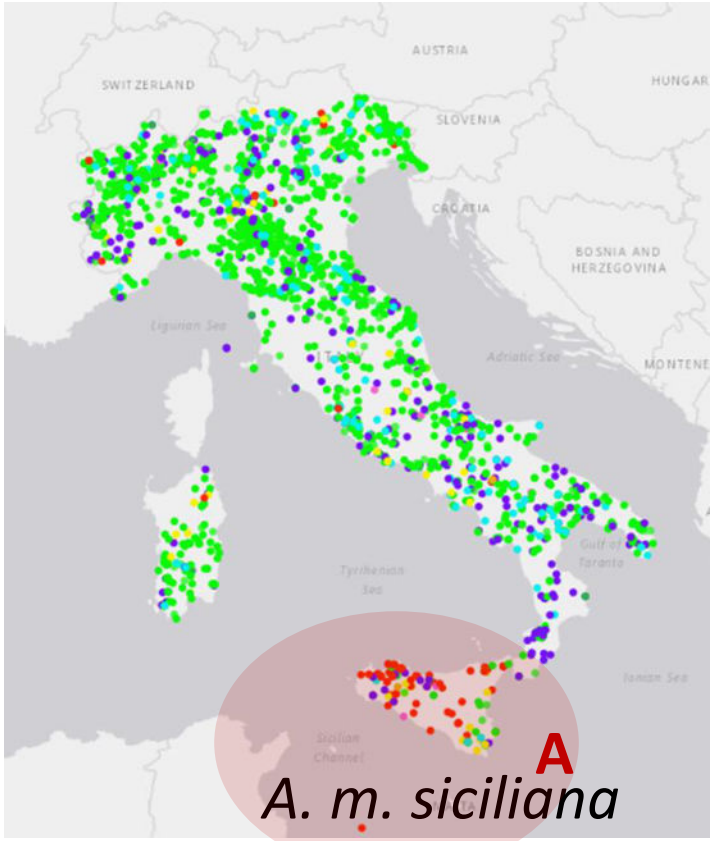


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Results

2018-2023 (all 6 years)

- C
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A, AC

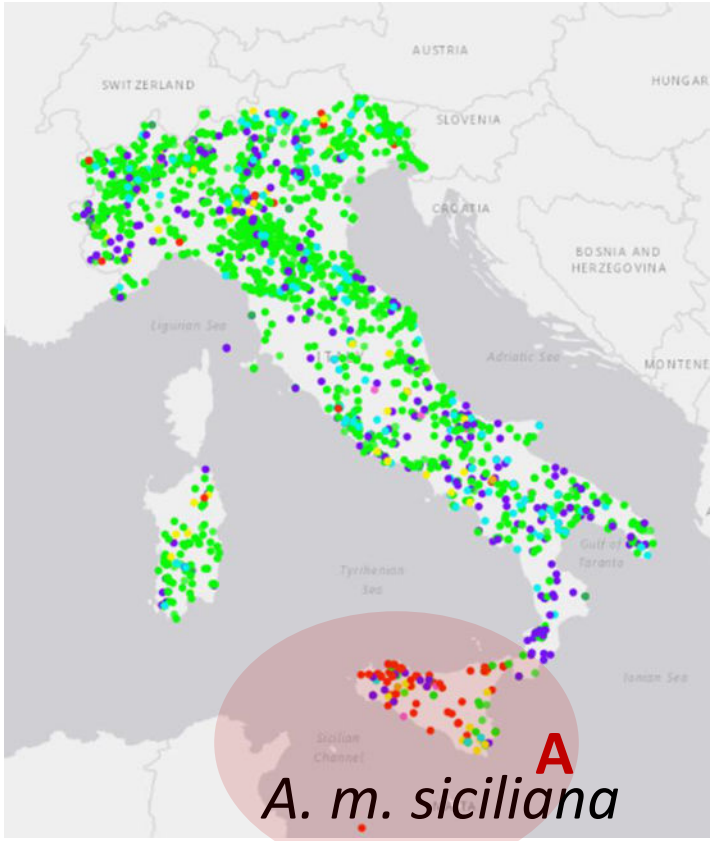


A, M, AC, AM, MC, AMC

Results

2018-2023 (all 6 years)

- C
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- M
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- MC
- AMC

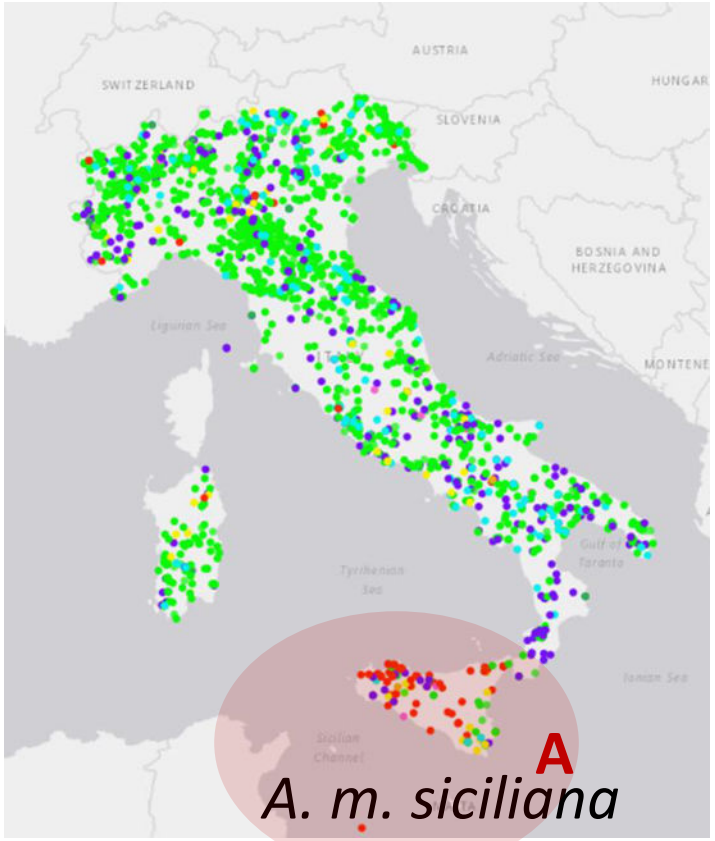




Results

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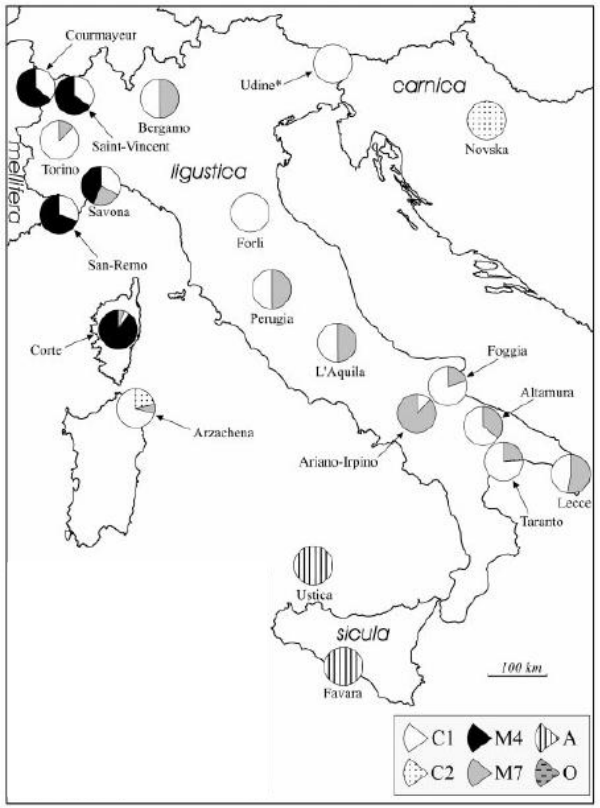


Molecular Ecology (2000) 9, 907–921

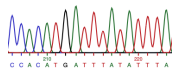
Hybrid origins of honeybees from Italy (*Apis mellifera ligustica*) and Sicily (*A. m. sicula*)

P. FRANCK,* L. GARNERY,† G. CELEBRANO,‡ M. SOLIGNAC† and J.-M. CORNUET*
*Centre de Biologie et de Gestion des Populations, Campus International de Baillarguet, 34980 Monferrier-sur-Lez, France,
†Laboratoire Populations, Génétique et Evolution, CNRS, Avenue de la Terrasse, 91198 Gif-sur-Yvette cedex, France,
‡Dipartimento di Biologia Animale, 17 via Accademia Albertina, 10123 Torino, Italy

2000



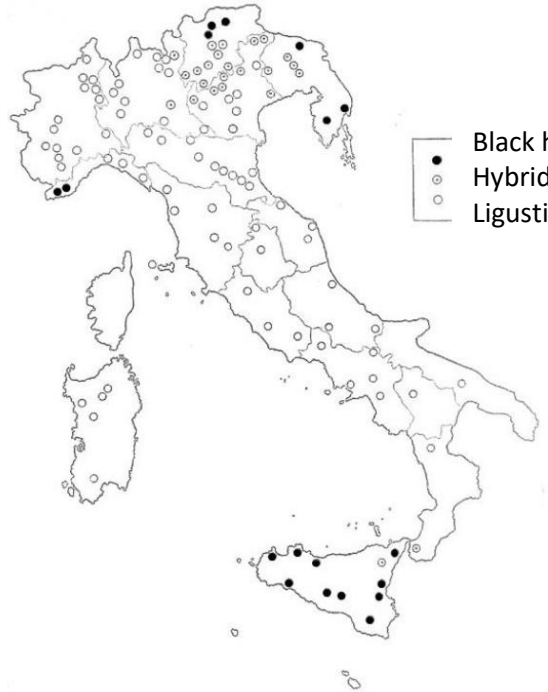






VECCHI A., 1927.- Sulla distribuzione geografica dell'*Apis mellifica ligustica* Spin. in Italia.- Bollettino del Laboratorio di zoologia generale e agraria della R. Scuola superiore d'agricoltura in Portici, 20: 150-168.

1927





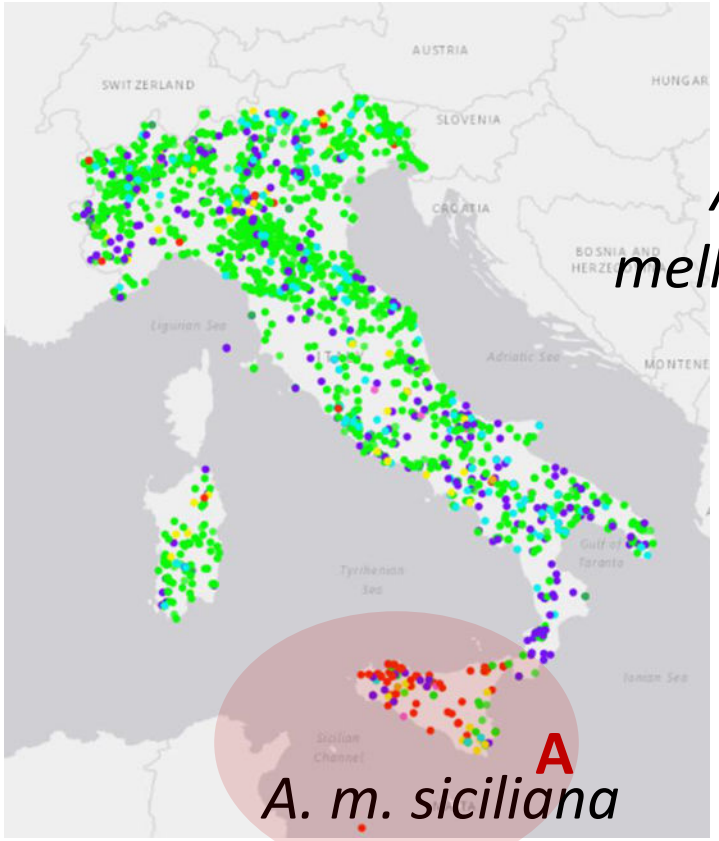


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Results

2018-2023 (all 6 years)

- C
- A
- M
- AC
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- MC
- AMC



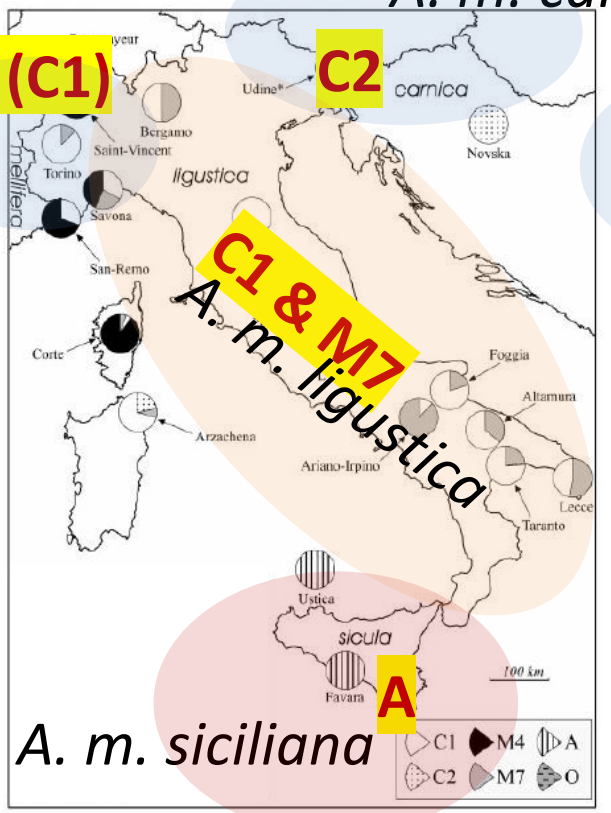


Molecular Ecology (2000) 9, 907–921

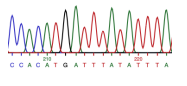
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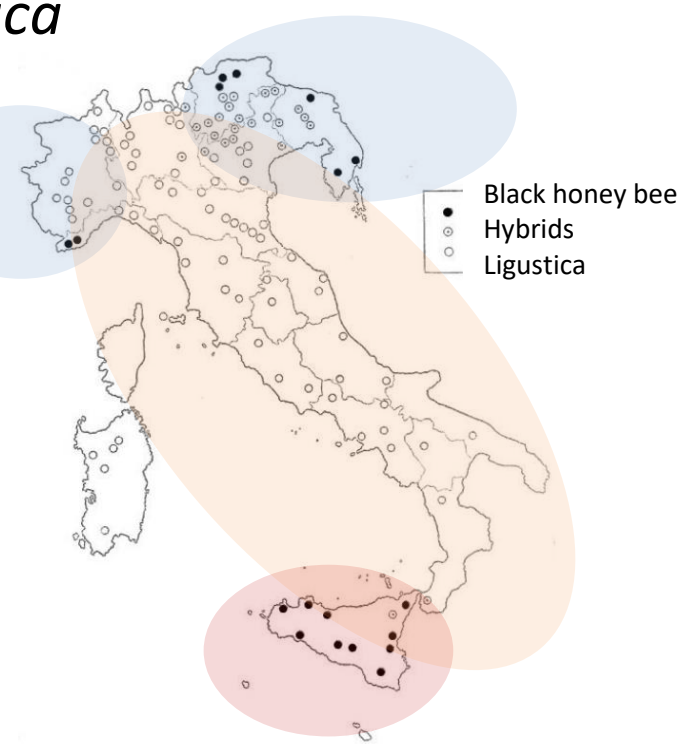






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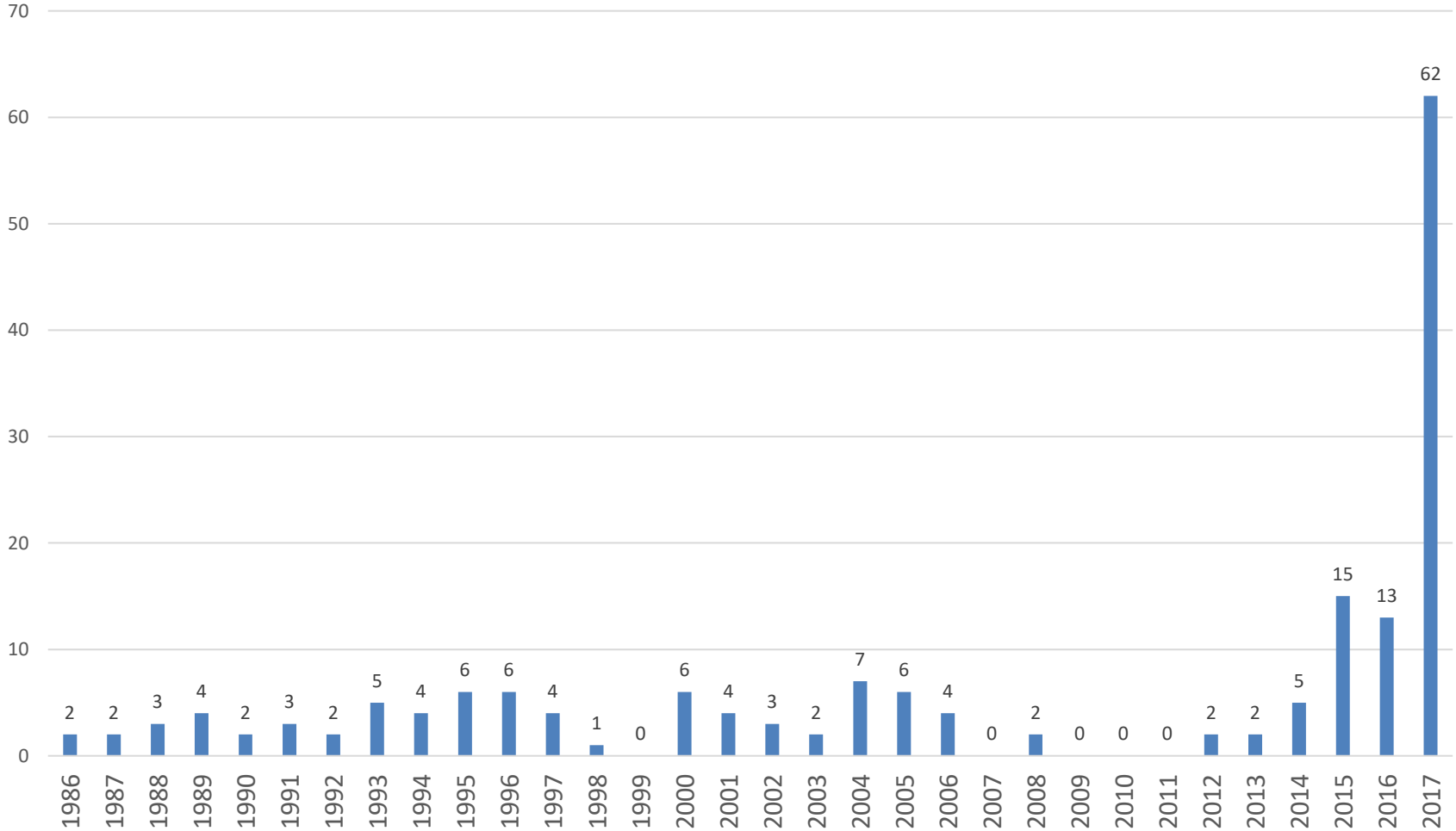


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Old samples (1986-2017)



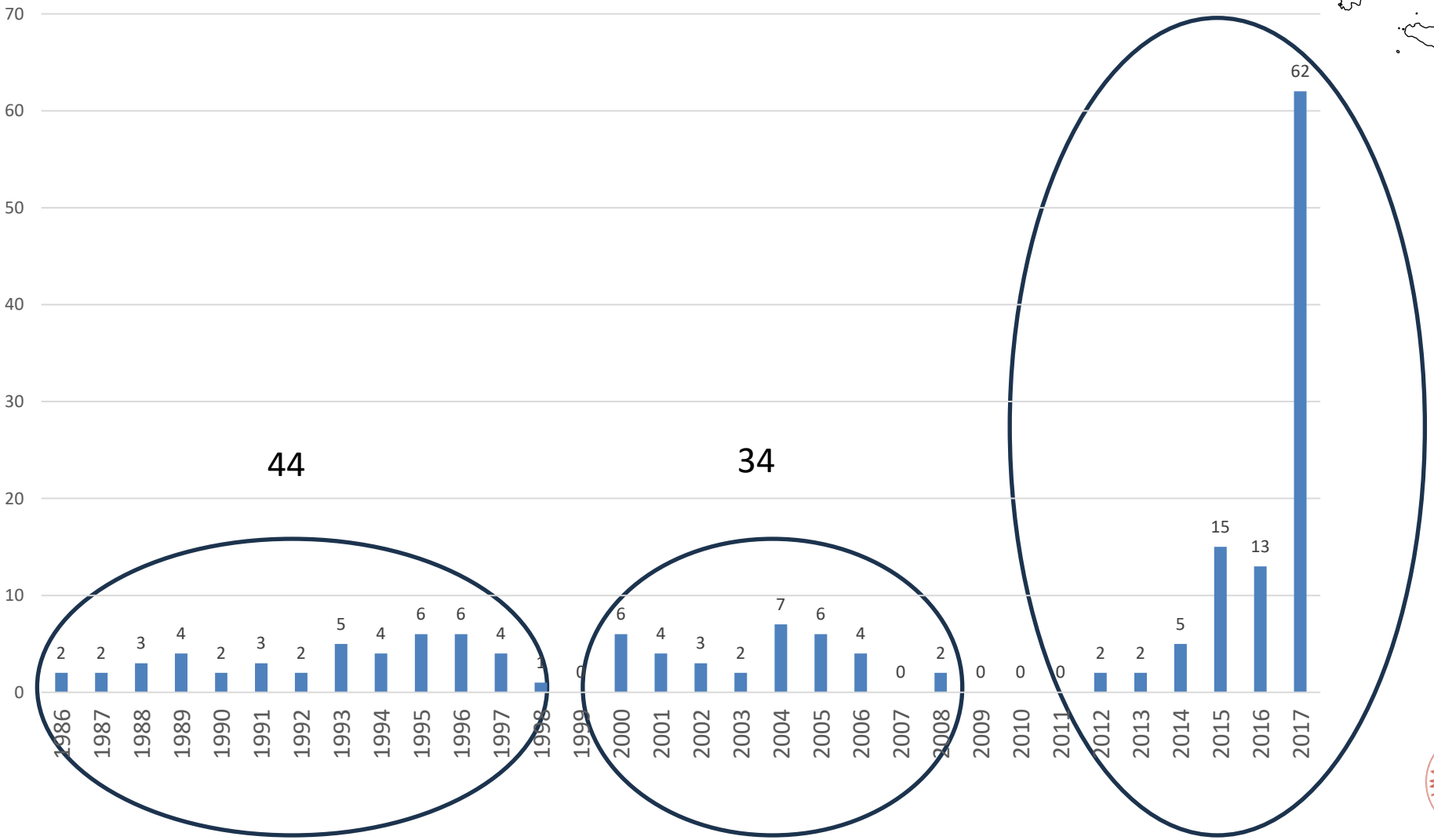


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Old samples (1986-2017)





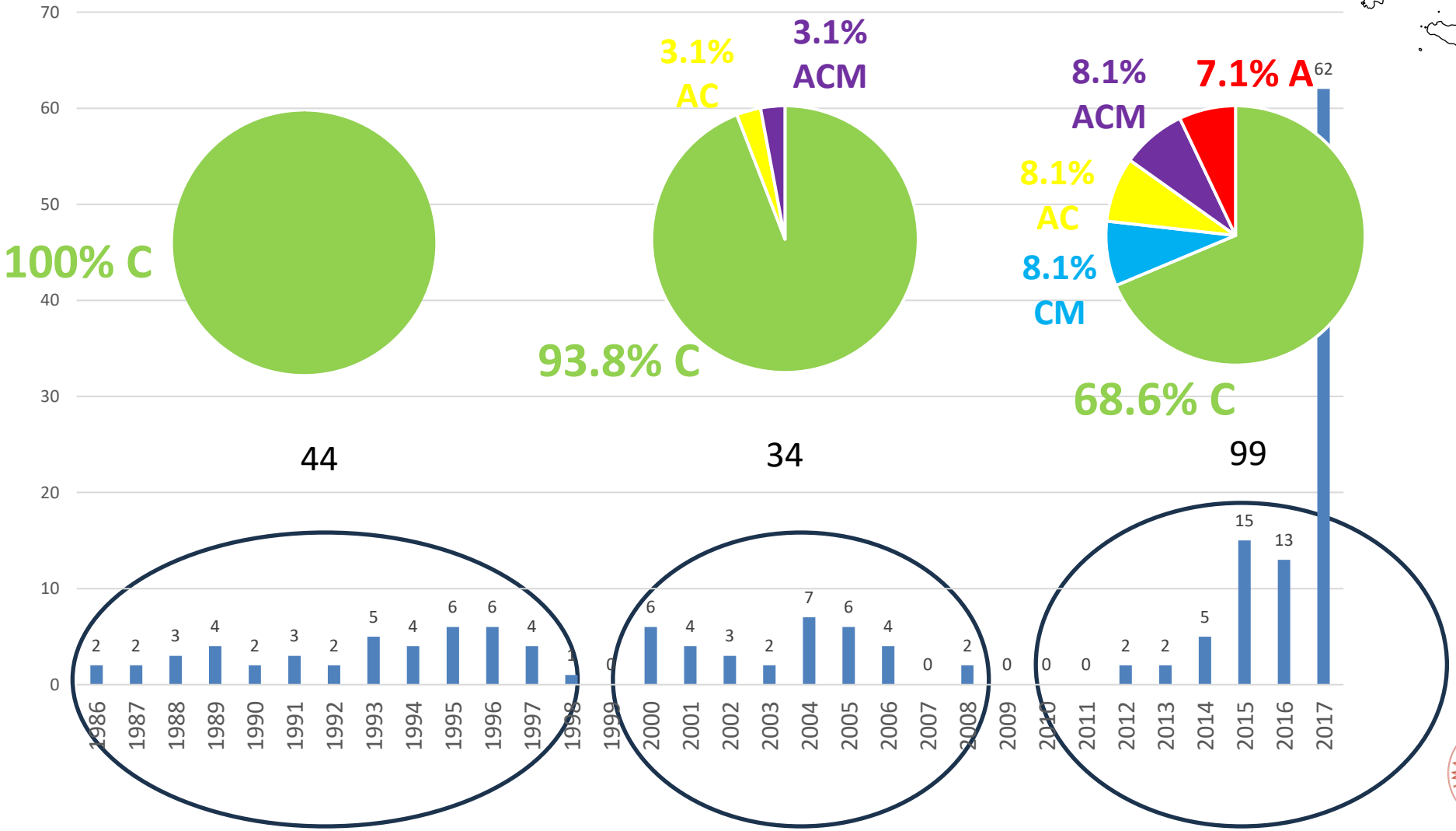
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Results





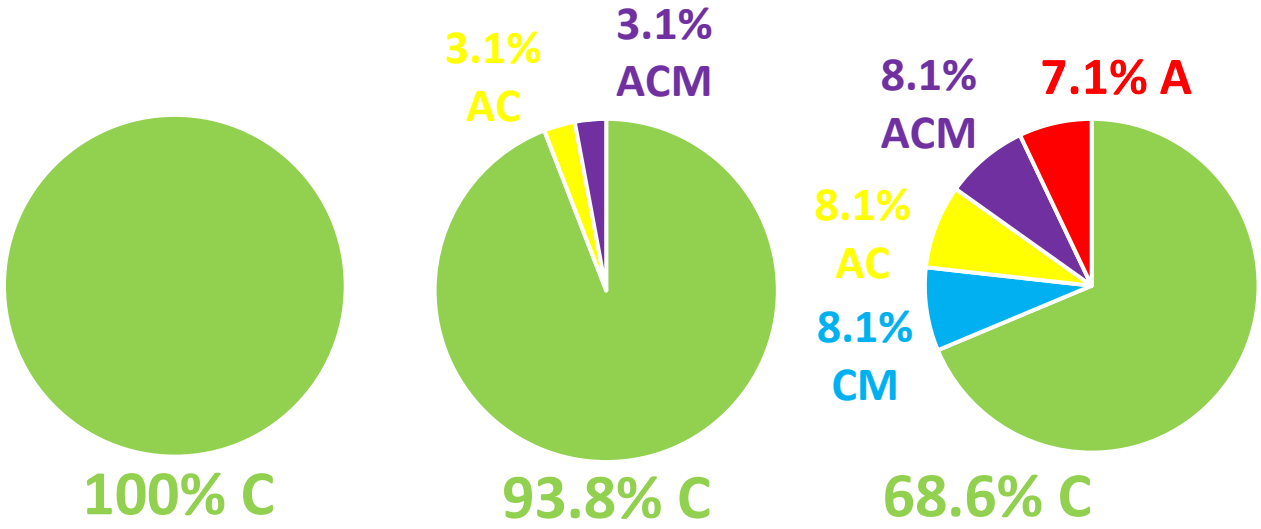
Old samples (1986-2017)



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Results





1986-1999

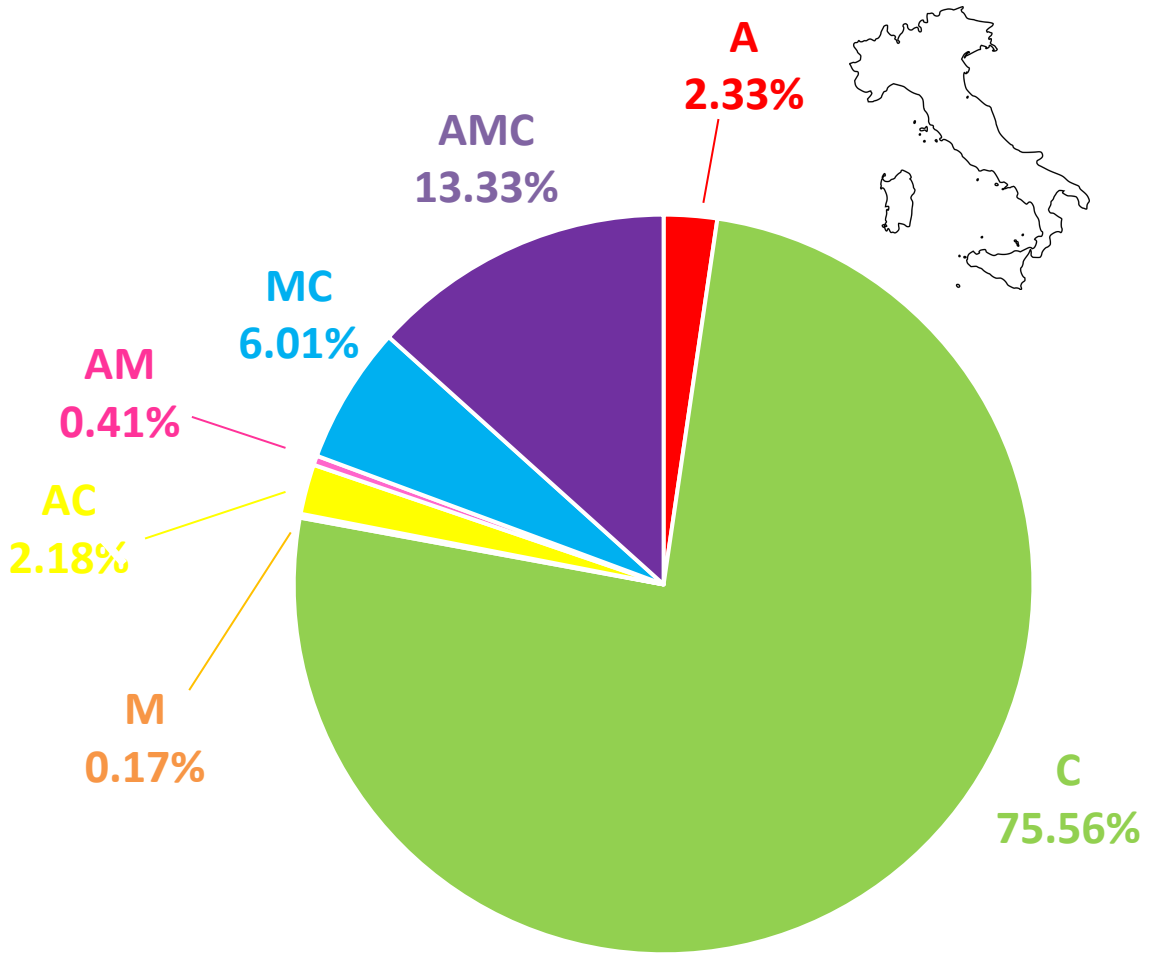
3

2000-2010

2

2011-2017

1



2018-2023

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2018



2019



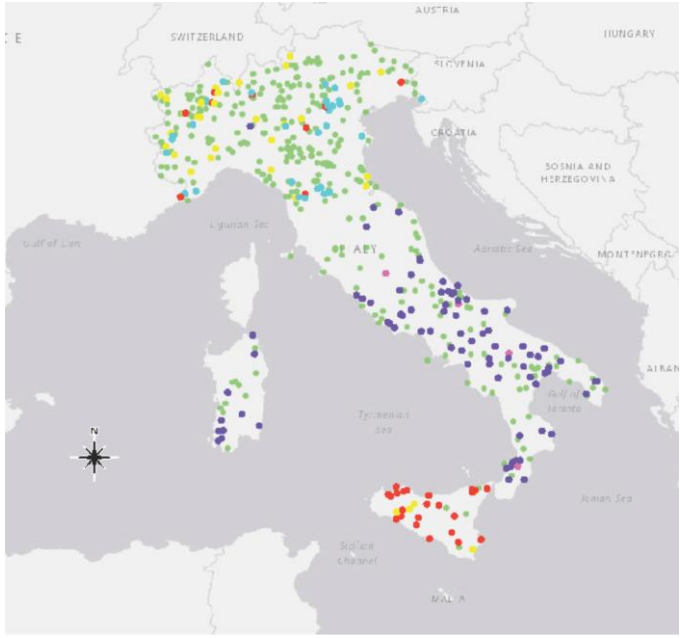
2020



2021



2022



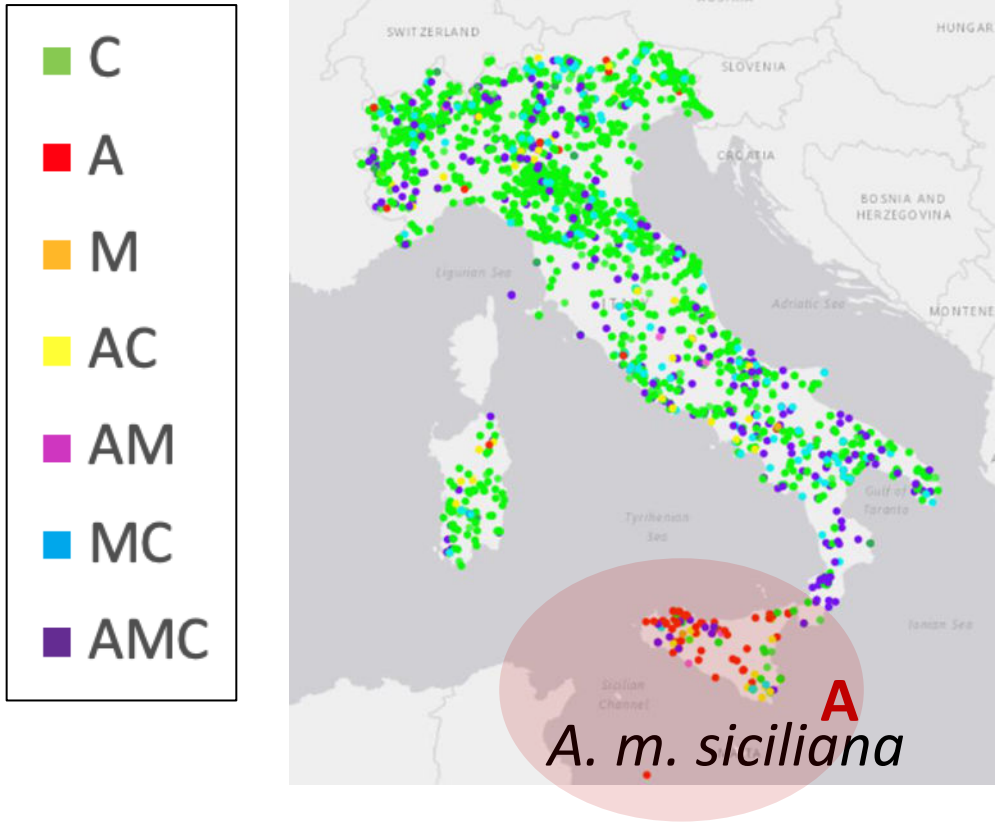
2023



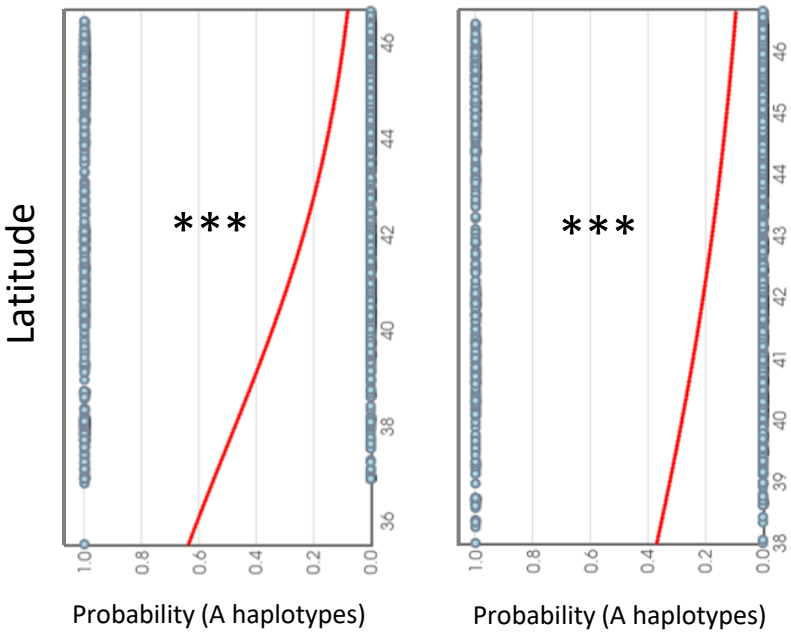
ALMA MATER STUDIORUM
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Results

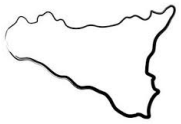
2018-2023 (all 6 years)



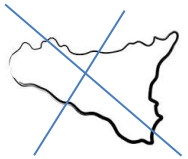
Logistic regression: all **A** honey samples



Including Sicily



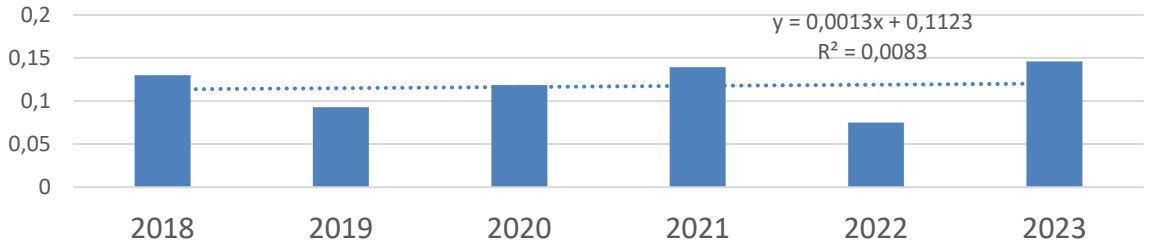
Excluding Sicily

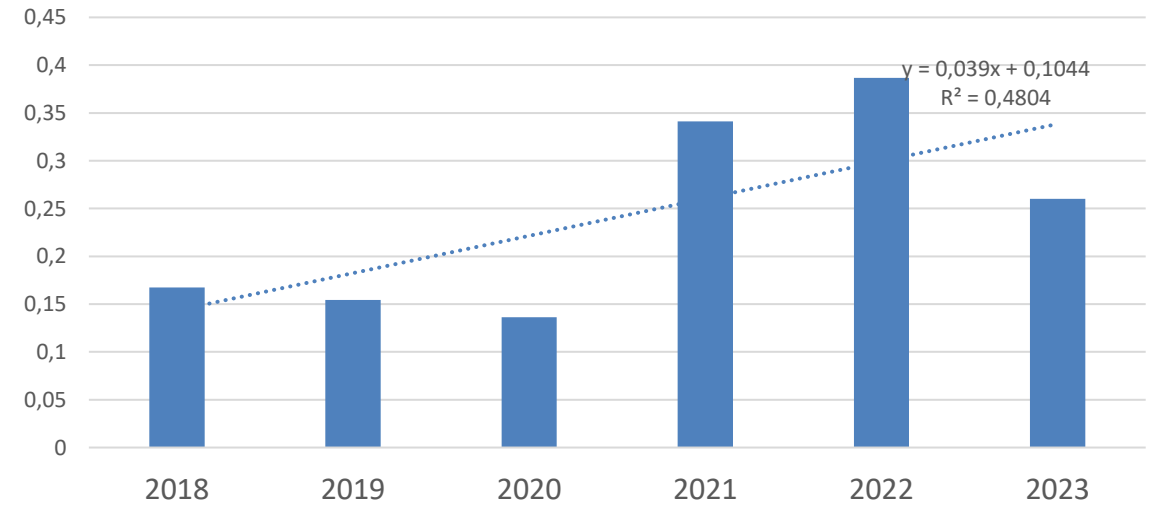


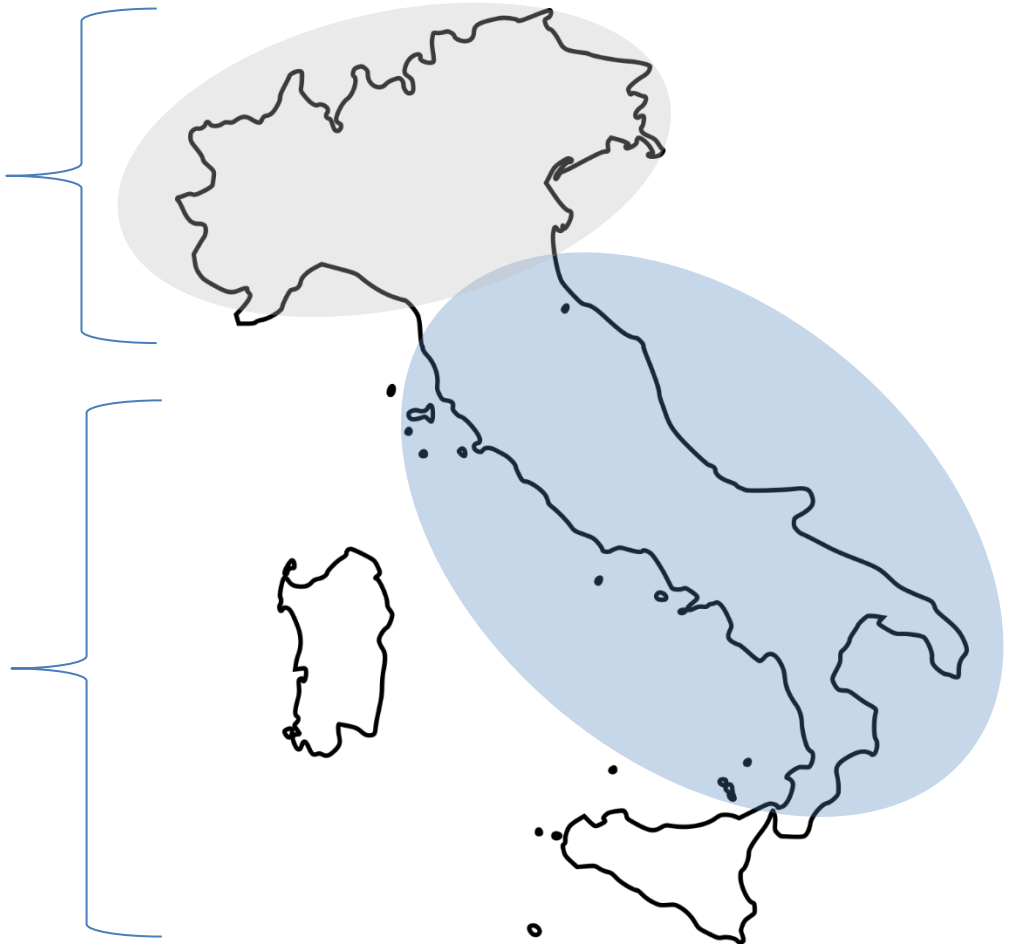
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Results

Honey samples with **A** mtDNA



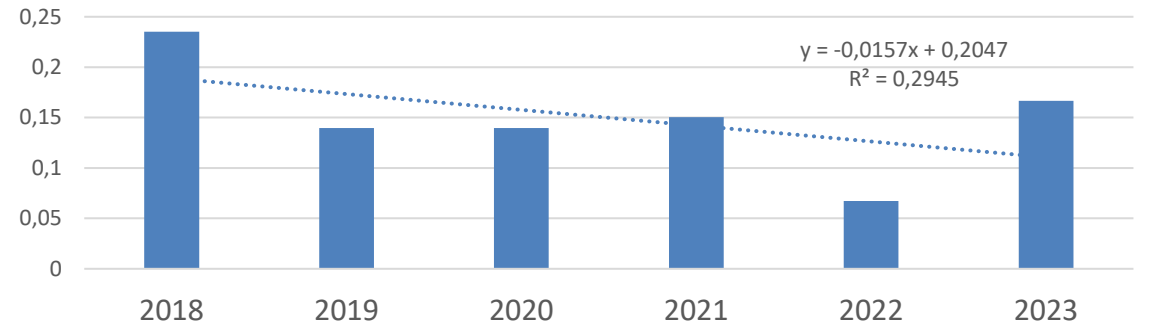


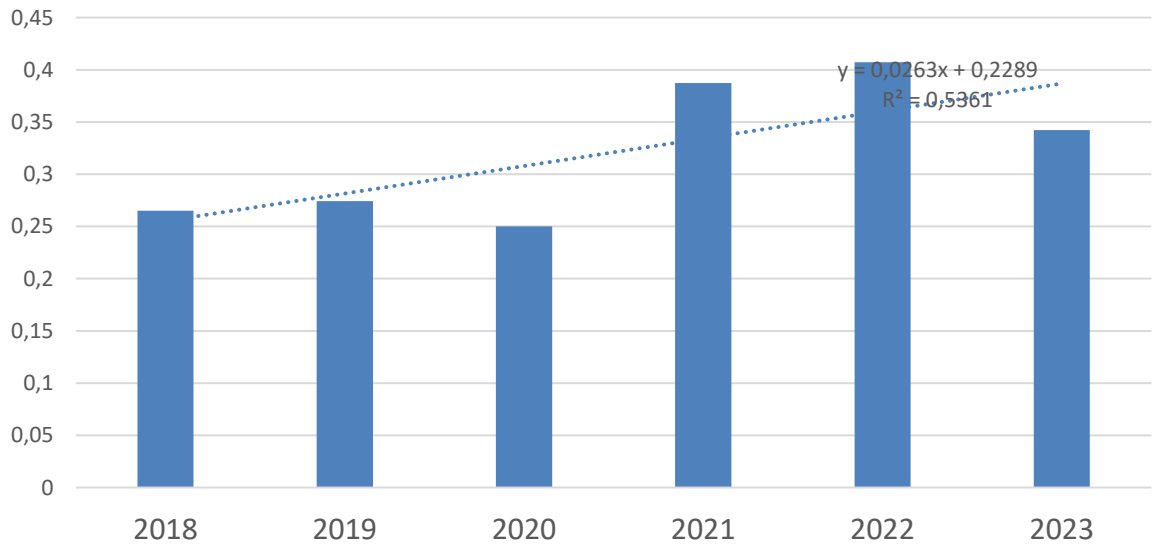


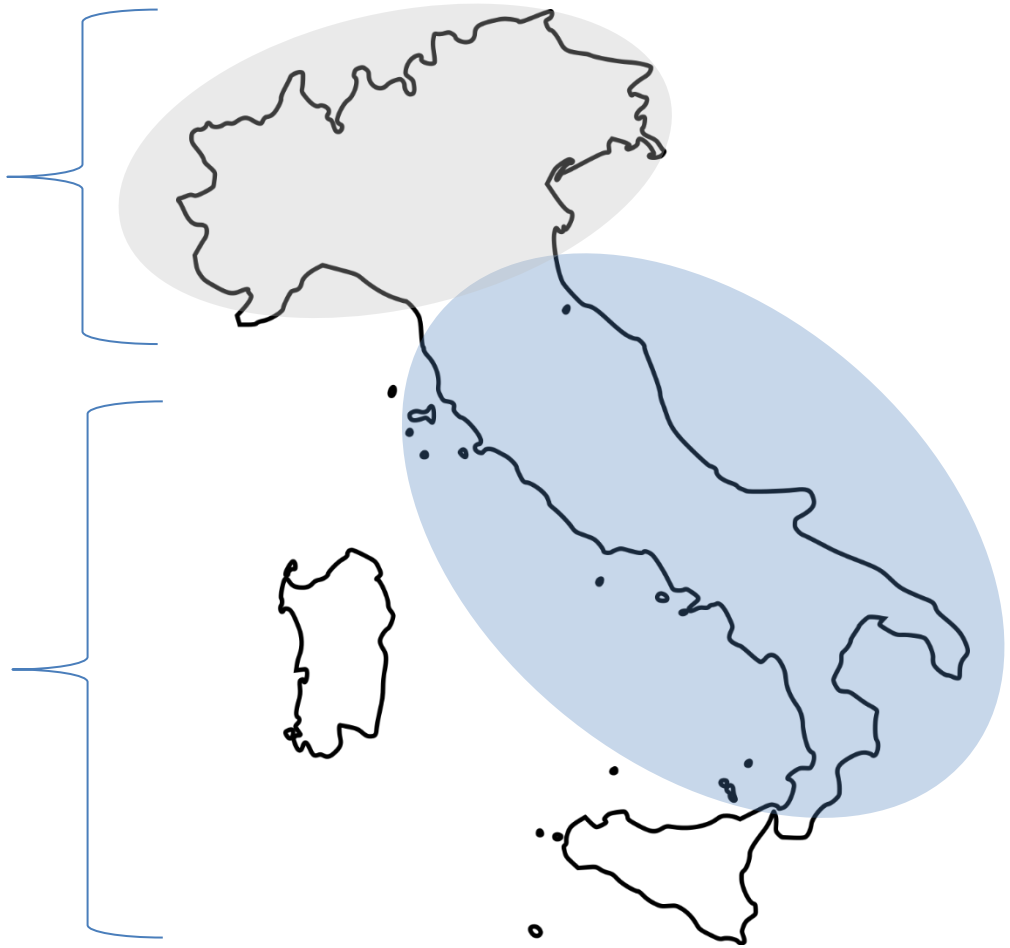
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Results

Honey samples with **M** mtDNA







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Validation

www.nature.com/scientificreports

scientific reports

Check for updates

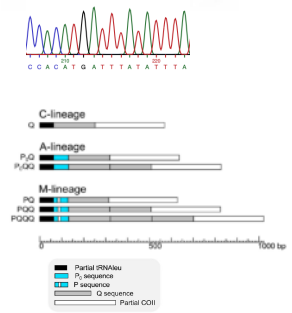
OPEN

Distribution of honey bee mitochondrial DNA haplotypes in an Italian region where a legislative act is protecting the *Apis mellifera ligustica* subspecies

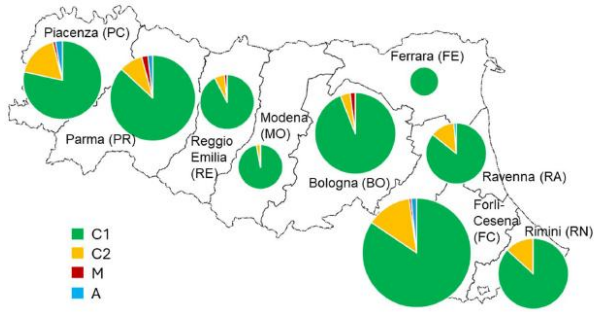
Valeria Taurisano, Anisa Ribani, Dalal Sami, Kate Elise Nelson Johnson, Giuseppina Schiavo, Valerio Joe Utzeri, Samuele Bovo & Luca Fontanesi✉

1143





2020-2022





A = 1.3%

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Validation

www.nature.com/scientificreports

scientific reports

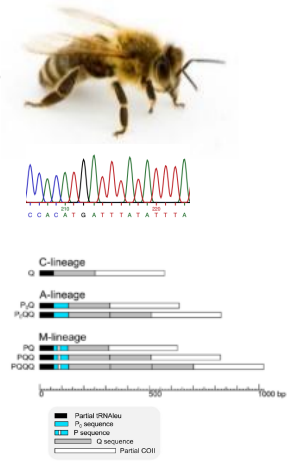
Check for updates

OPEN

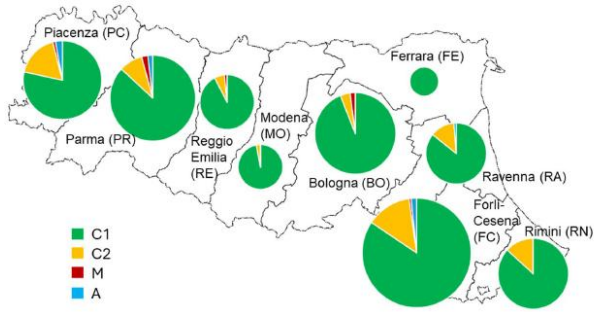
Distribution of honey bee mitochondrial DNA haplotypes in an Italian region where a legislative act is protecting the *Apis mellifera ligustica* subspecies

Valeria Taurisano, Anisa Ribani, Dalal Sami, Kate Elise Nelson Johnson, Giuseppina Schiavo, Valerio Joe Utzeri, Samuele Bovo & Luca Fontanesi✉

1143



2020-2022





A = 1.3%

429



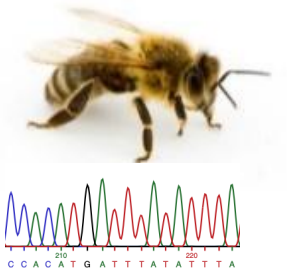
2020-2022



ACM + AC = 4.7%

Final considerations

1



≈

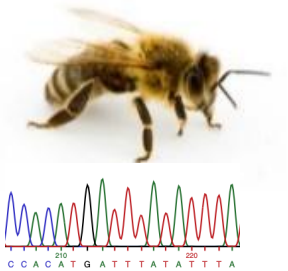


for honey bee population genetic studies

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Final considerations

1



≈



for honey bee population genetic studies

2

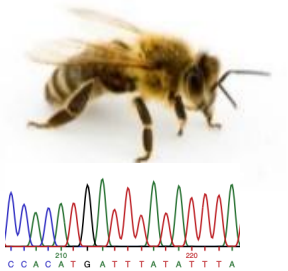


can be more effective for large scale population genetic studies (1-2 orders of magnitute larger)

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Final considerations

1



≈



for honey bee population genetic studies

2



can be more effective for large scale population genetic studies (1-2 orders of magnitute larger)

3

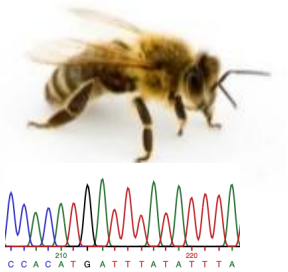


honey bee populations have been changed over the last decades and are still changing (introduction of «alien» mtDNA haplotypes)

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Final considerations

1



≈



for honey bee population genetic studies

2



can be more effective for large scale population genetic studies (1-2 orders of magnitute larger)

3



honey bee populations have been changed over the last decades and are still changing (introduction of «alien» mtDNA haplotypes)

4



What are the implications of this «modification» ?

11th European Conference of Apidology

7-10 September 2026 · Bologna, Italy

LIKE FORAGING BEES, WE SEEK NOVELTY, WE NEED COOPERATION AND WE PROFIT FROM DIVERSITY

The conference is jointly organized by CREA-AA – Research Centre for Agriculture and Environment and the Alma Mater Studiorum – University of Bologna

EurBee11
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Acknowledgments







OSSERVATORIO NAZIONALE MIELE

piana RICERCA E CONSULENZA

Afapicoltori

ASSOCIAZIONE APICOLTORI REGGIO PARMA

ASSOCIAZIONE ROMAGNOLA APICOLTORI

ASSOCIAZIONE PROVINCIALE APICOLTORI PIACENTINI

ItaHoneyDNA

MINISTERO DELL'AGRICOLTURA DELLA SOVRANITÀ ALIMENTARE E DELLE FORESTE

de nostre api

Associazione Apicoltori Emilia-Romagna



MIELE IN COOPERATIVA

FAI FEDERAZIONE APICOLTORI ITALIANI

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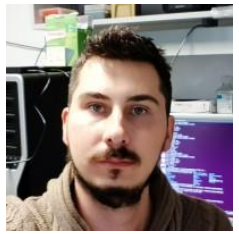
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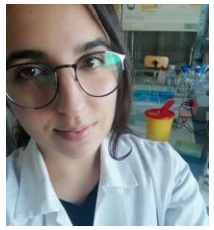
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https://site.unibo.it/animal-and-food-genomics/en

AFG Group

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Animal and Food Genomics Group

A dynamic picture of the diffusion of *Apis mellifera* mitochondrial DNA lineages in Italy over the last few decades utilizing honey as a source of honey bee population genetic information



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