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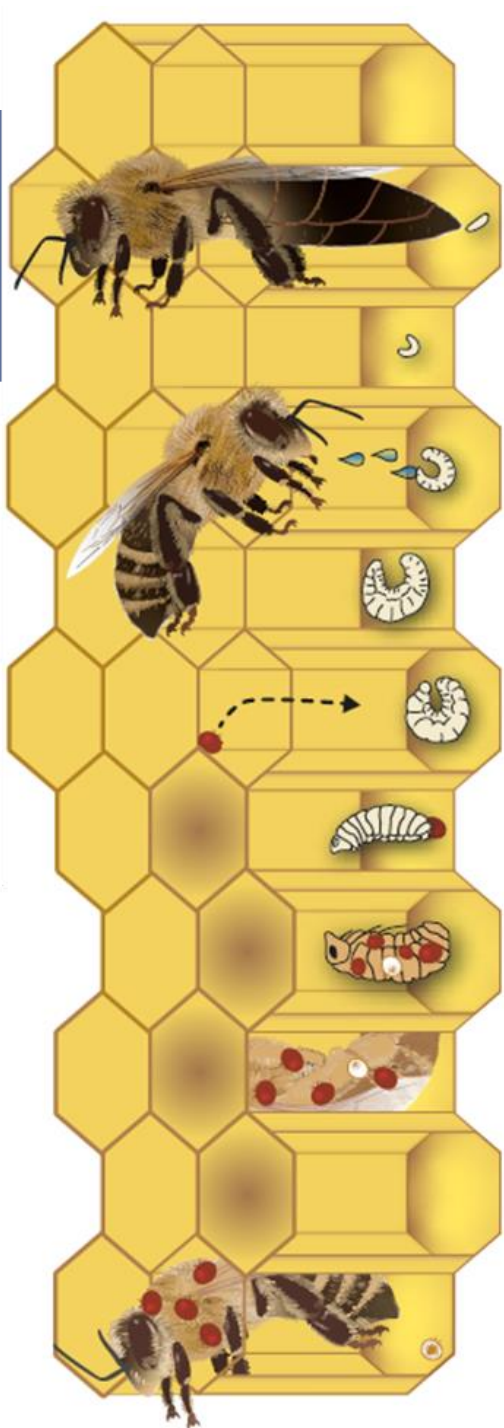
Varroa destructor still reflects a major threat to honey bees and beekeeping around the globe. This key role for apiculture keeps it in focus of numerous breeding attempts aiming to select varroa-resistant stocks. However, the exact pathways of varroa-resistance are still largely unknown, which greatly hampers the selection of corresponding traits. We studied the effects of outer factors on the reproductive failure of mites ("MNR"), the recapping behaviour of bees ("REC"), the interaction of these traits and their applicability in breeding. To this end, we first investigated MNR in colonies with brood interruptions of varying durations ($n = 27$). In addition, we repeatedly sampled (five times per year) colonies over the course of three years ($n = 20, 20$ and 15 , respectively) to investigate temporal variation in the accounted traits. Finally, the heritability (h^2) of both traits was calculated based on an extensive dataset of 1784 Buckfast and 2530 Carnica colonies. Colonies with interrupted brood rearing showed higher MNR (GLMM: $\chi^2 = 15.2$, $p < 0.001$, $df = 1$) and REC ($\chi^2 = 8.09$, $p = 0.005$, $df = 1$) values in comparison to broodright colonies. Both traits were also strongly affected by seasonal variation (MNR: $\chi^2 = 152.23$; REC: $\chi^2 = 335.5$; each: $p < 0.001$, $df = 14$; $n = 4106$ single-infested cells). In addition, REC was associated with certain forms of MNR, i.e., delayed reproduction ($\chi^2 = 9.15$, $p = 0.003$, $df = 1$) and missing males ($\chi^2 = 8.1$, $p = 0.004$, $df = 1$). Recapping activity was also affected by brood age ($\chi^2 = 238.13$, $p < 0.001$, $df = 4$) and most frequent at day 6 post-capping, i.e., when the two eldest descendants would be expected (Tukey: $p < 0.05$; 764 recapped cells). With an accessible h^2 of 0.18 and 0.44 for MNR and 0.44 and 0.40 for REC, both traits proved promising for selection in Buckfast and Carnica stocks, respectively. A more detailed calculation including apiary factors for Carnica yielded even higher h^2 (0.51 & 0.40, $n = 2233$ & 2114 colonies for MNR and REC, respectively). Implications of these findings for practical breeding will be discussed.

Chances and challenges in breeding for resistance to *Varroa destructor* – from basic principles to practical application

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Apimondia Copenhagen,
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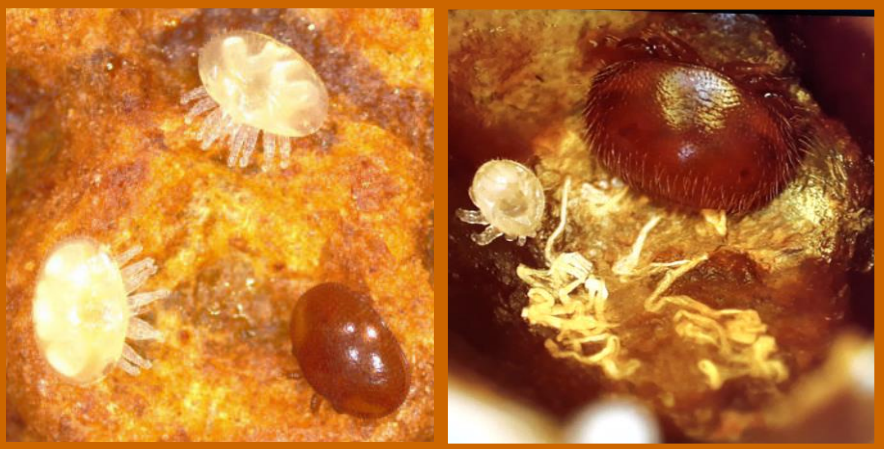
MNR
mite non-reproduction



male missing

delayed

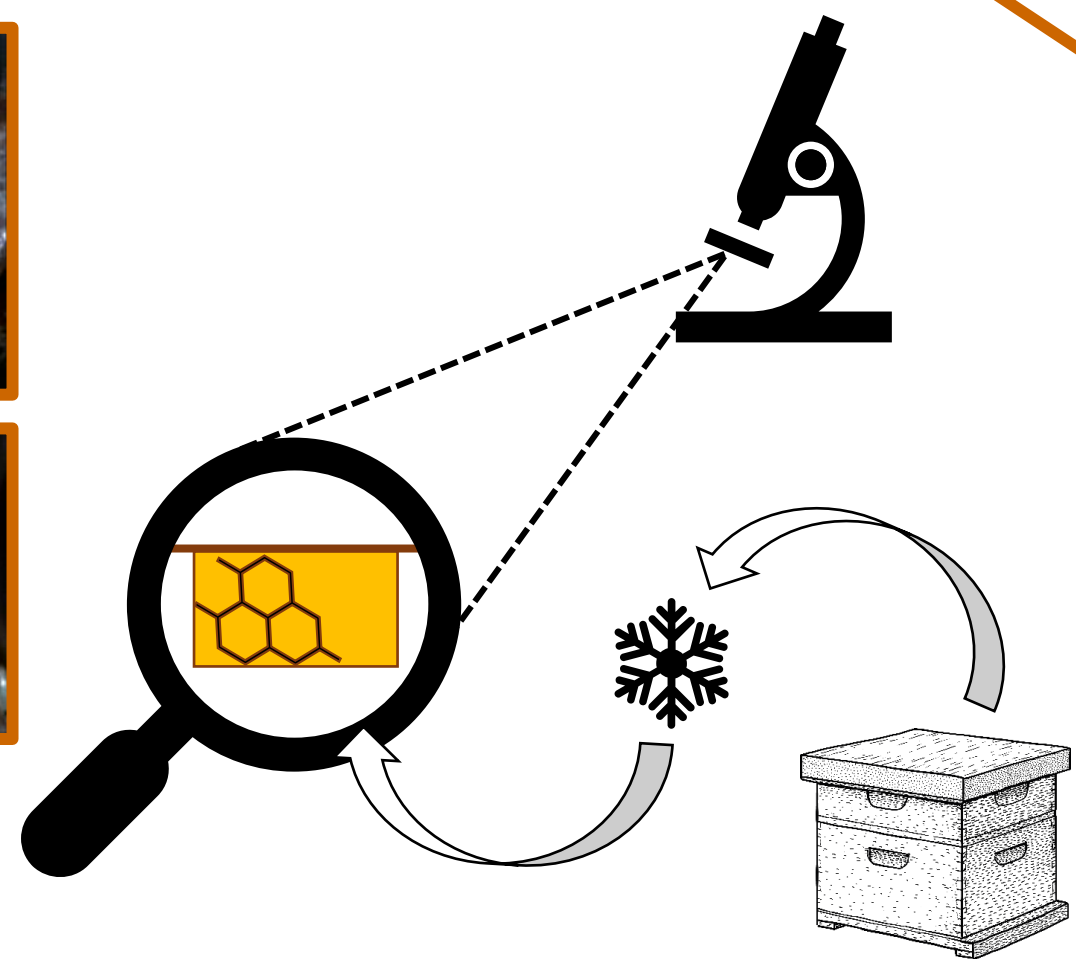
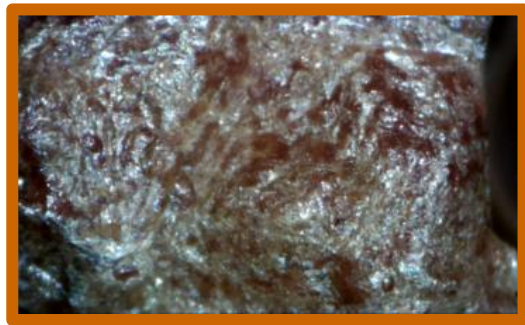
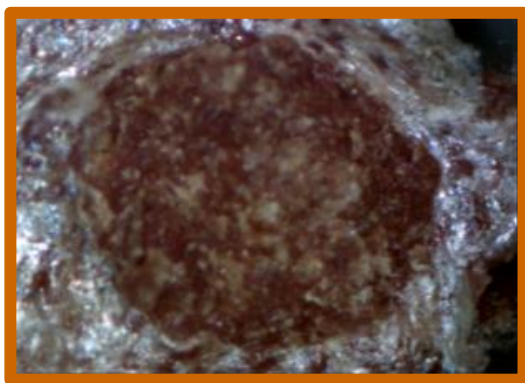
infertile

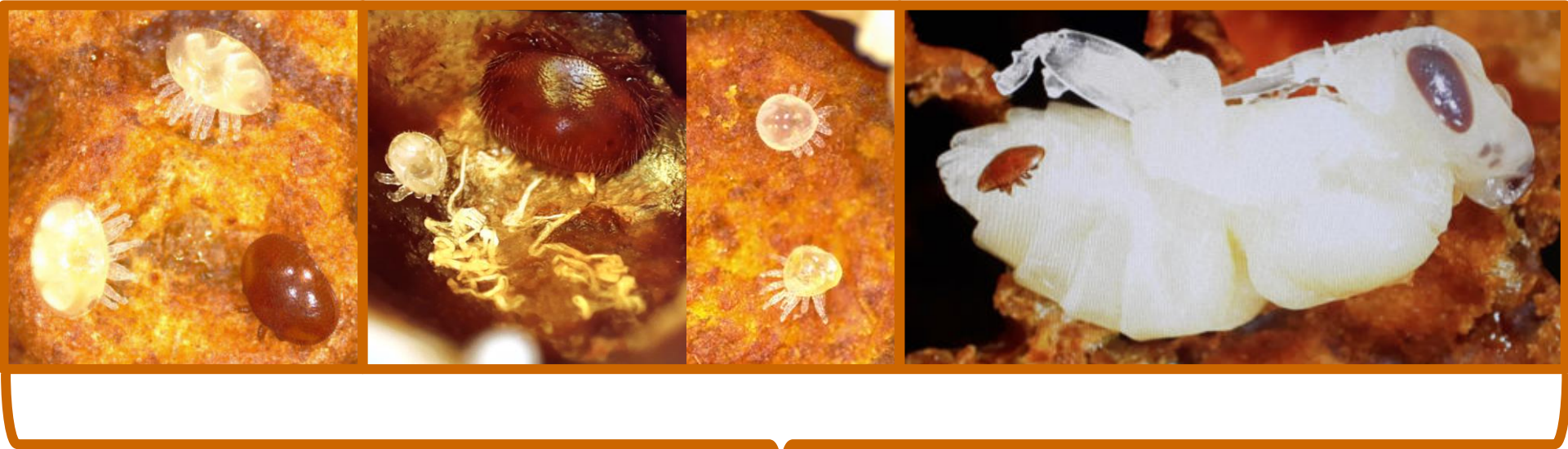


MNR
mite non-reproduction



REC
uncapping & recapping
of broodcells





From basics to application:

- **Identify mechanisms** that lead to decreased mite reproduction
- Investigate **effects of recapping** behaviour on cell level
- **Draw implications** for resistance breeding and beekeeping practice

MNR

mite non-reproduction

REC

uncapping & recapping
of broodcells

**other
factors**





other
factors



**Effect of Brood Interruptions
on MNR**
mite non-reproduction

Hive
management

Honey bee
biology

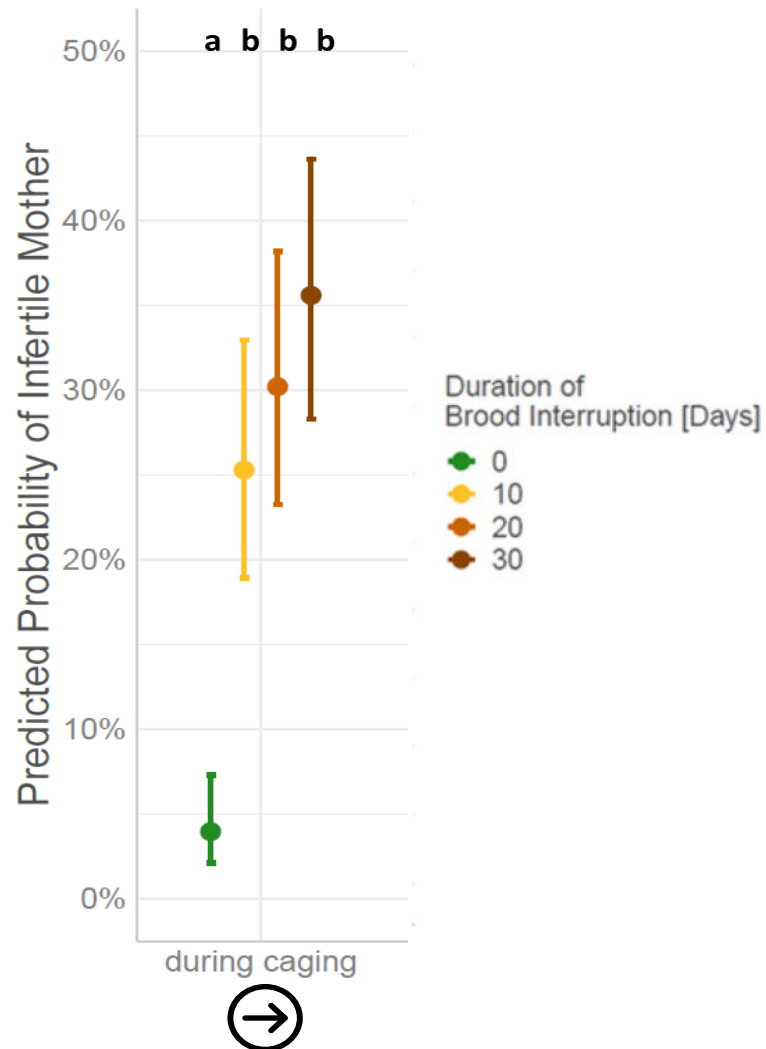
Bioassays used
in research &
breeding

Effects of brood interruptions on the reproductive success of *Varroa destructor*

- 27 full grown colonies
 - Same breeding stock
 - Common apiary
- Induced brood interruptions by queen caging
 - 10, 20 or 30 days (n = 7, each)
 - Control group with continuous brood rearing (n = 6)
- Sampling during caging and in the first, second and third brood cycle afterwards

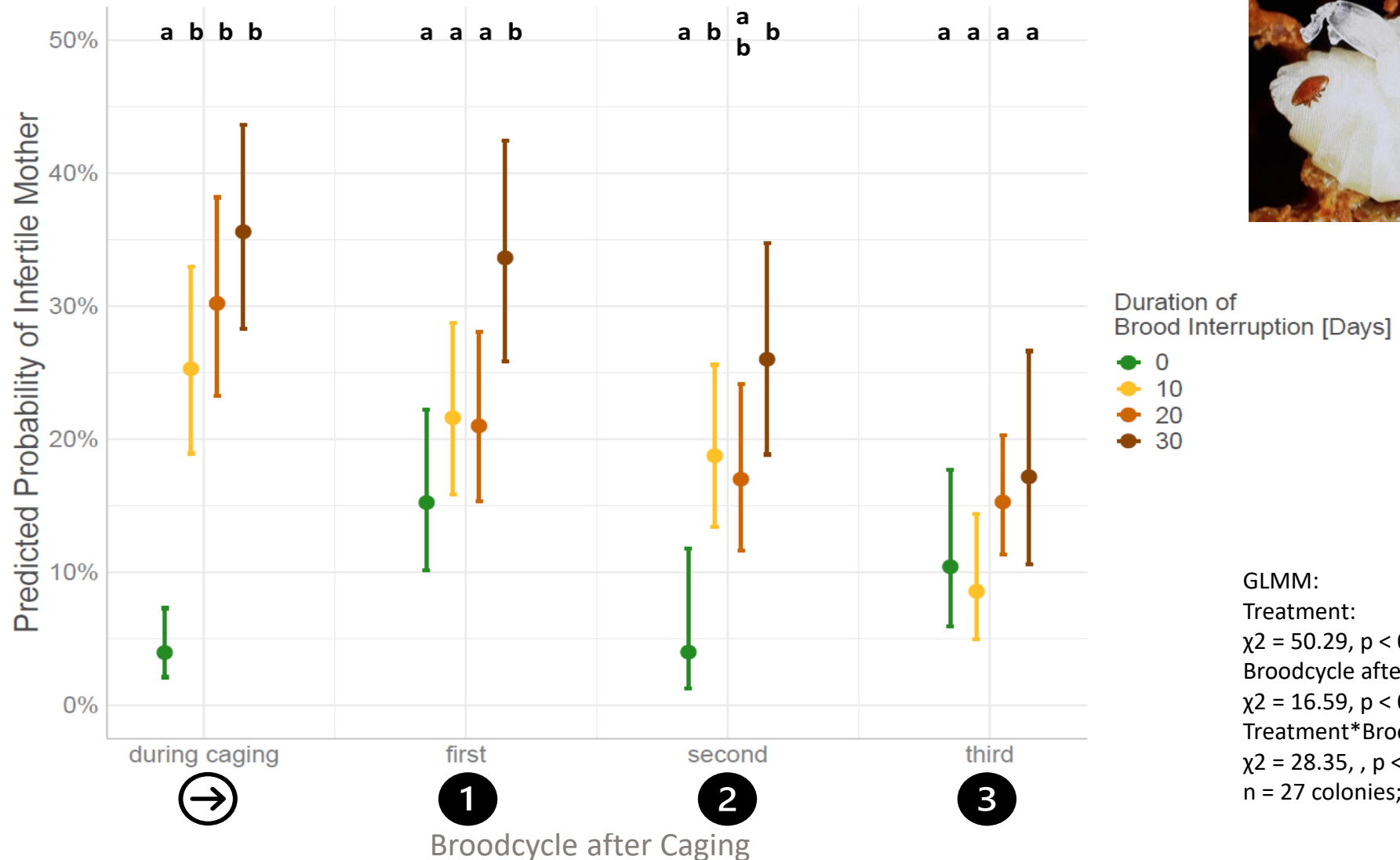


Brood interruptions increase the occurrence of infertile mites immediately

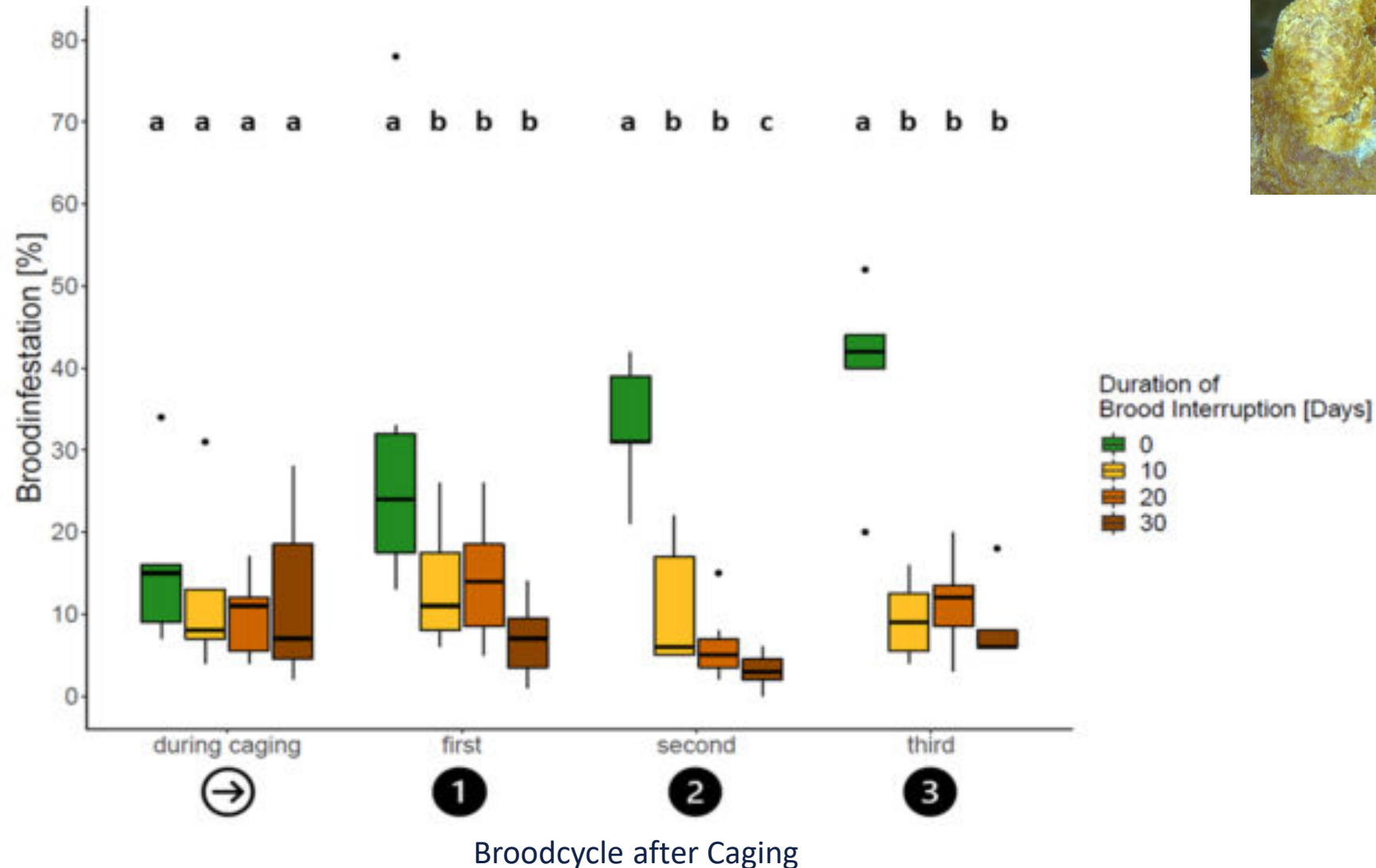


GLMM:
 Treatment:
 $\chi^2 = 50.29$, $p < 0.001$, $df = 3$;
 Broodcycle after Caging:
 $\chi^2 = 16.59$, $p < 0.001$, $df = 3$;
 Treatment*Broodcycle after Caging:
 $\chi^2 = 28.35$, $p < 0.001$, $df = 9$;
 $n = 27$ colonies; 95% CI displayed

Brood interruptions increase the occurrence of infertile mites immediately and in the long-term



Brood interruptions result in a long-lasting decrease of *Varroa*-infestation



GLMM:
 Treatment:
 $\chi^2 = 27.42$, $p < 0.001$, $df = 3$;
 Broodcycle after Caging:
 $\chi^2 = 3.02$, $p = 0.03$, $df = 3$;
 Treatment*Broodcycle after Caging:
 $\chi^2 = 3.2$, $p < 0.001$, $df = 9$;
 $n = 27$ colonies; Boxplots display median values (horizontal line in box body), 1st & 3rd quartiles (box), minimum & maximum (whiskers) and outliers (dots)

other
factors



REC

uncapping & recapping
of broodcells



**Seasonal
Variation in MNR**
mite non-reproduction



Seasonal variation of mite non-reproduction and recapping

Long term study between 2019 and 2021

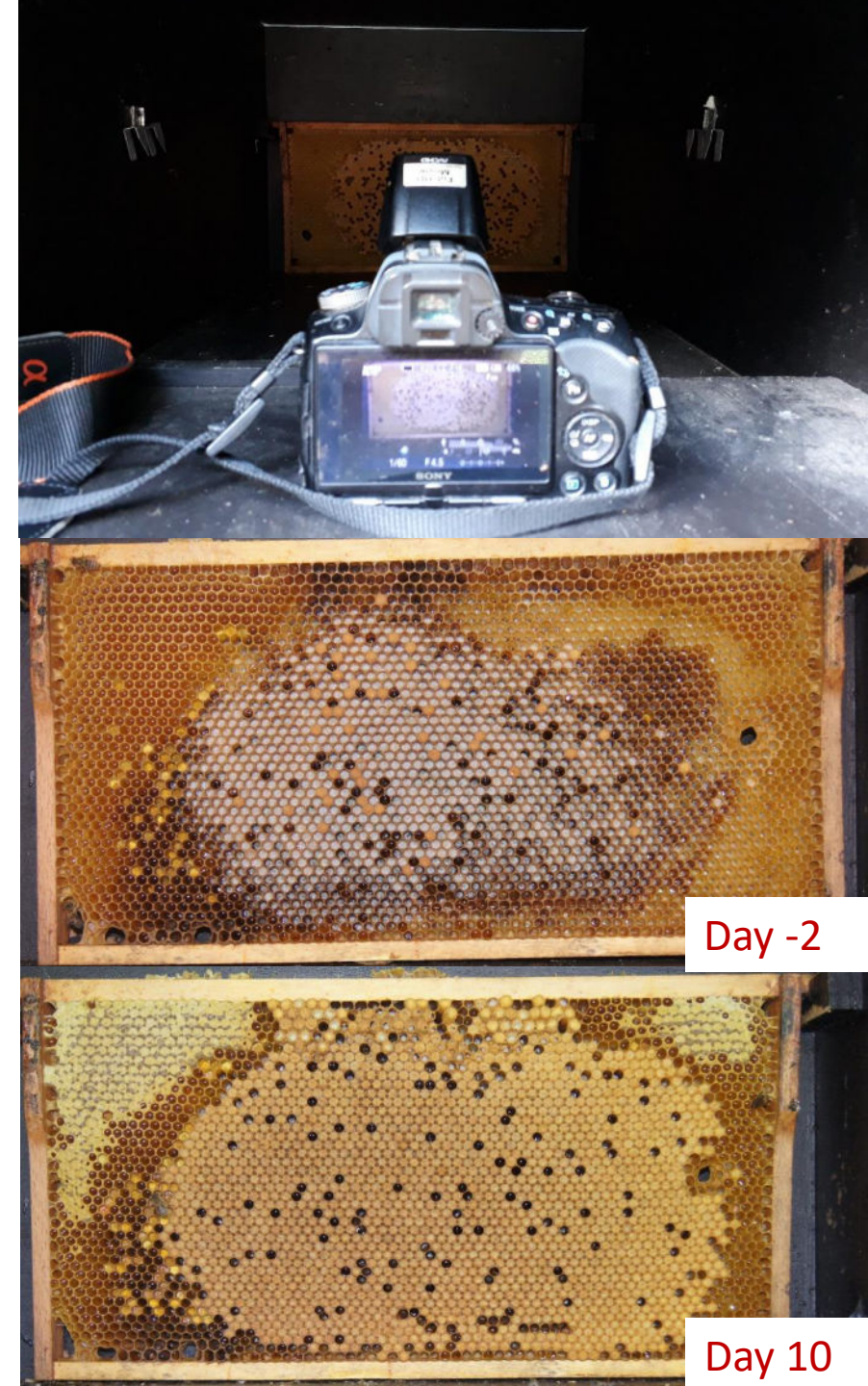
- 15 - 20 full grown colonies per year
 - Same breeding stock
 - Common apiary



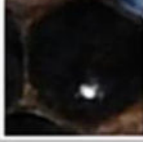
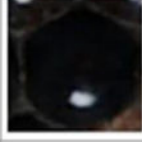
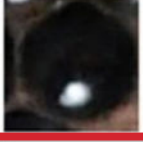
Seasonal variation of mite non-reproduction and recapping

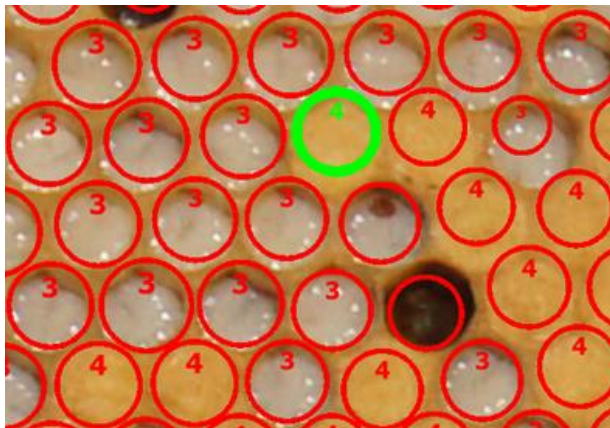
Long term study between 2019 and 2021

- 15 - 20 full grown colonies per year
 - Same breeding stock
 - Common apiary
- Repeated sampling throughout each season (5 trials)
 - Picture-based brood analysis
 - Brood samples for recapping and reproduction analysis
 - Bee samples for infestation measurements

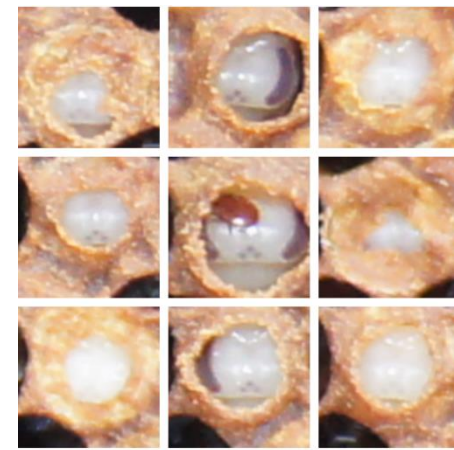


Picture-based brood analysis: Recapping and brood termination

ID	BFD 0	BFD 2	BFD 4	BFD 6	BFD 8	BFD 10	BFD 12
51	 Old Larva	 Pupa	 Pupa	 Pupa	 Pupa	 Pupa	 Pupa
50	 Old Larva	 Pupa	 Pupa	 Pupa	 REC	 Pupa	 Pupa
52	 Old Larva	 Pupa	 Pupa	 Empty	 Empty	 Nectar	 Nectar

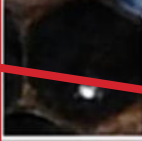
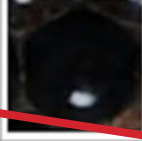
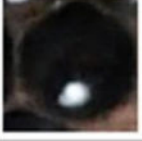


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ID	BFD 0		BFD 2		BFD 4		BFD 6		BFD 8		BFD 10		BFD 12	
51		Old Larva		Pupa		Pupa		Pupa		Pupa		Pupa		Pupa
50		Old Larva		Pupa		Pupa		Pupa		REC		Pupa		Pupa
52		Old Larva		Pupa		Pupa		Empty		Empty		Nectar		Nectar

Picture-based brood analysis: Recapping and brood termination

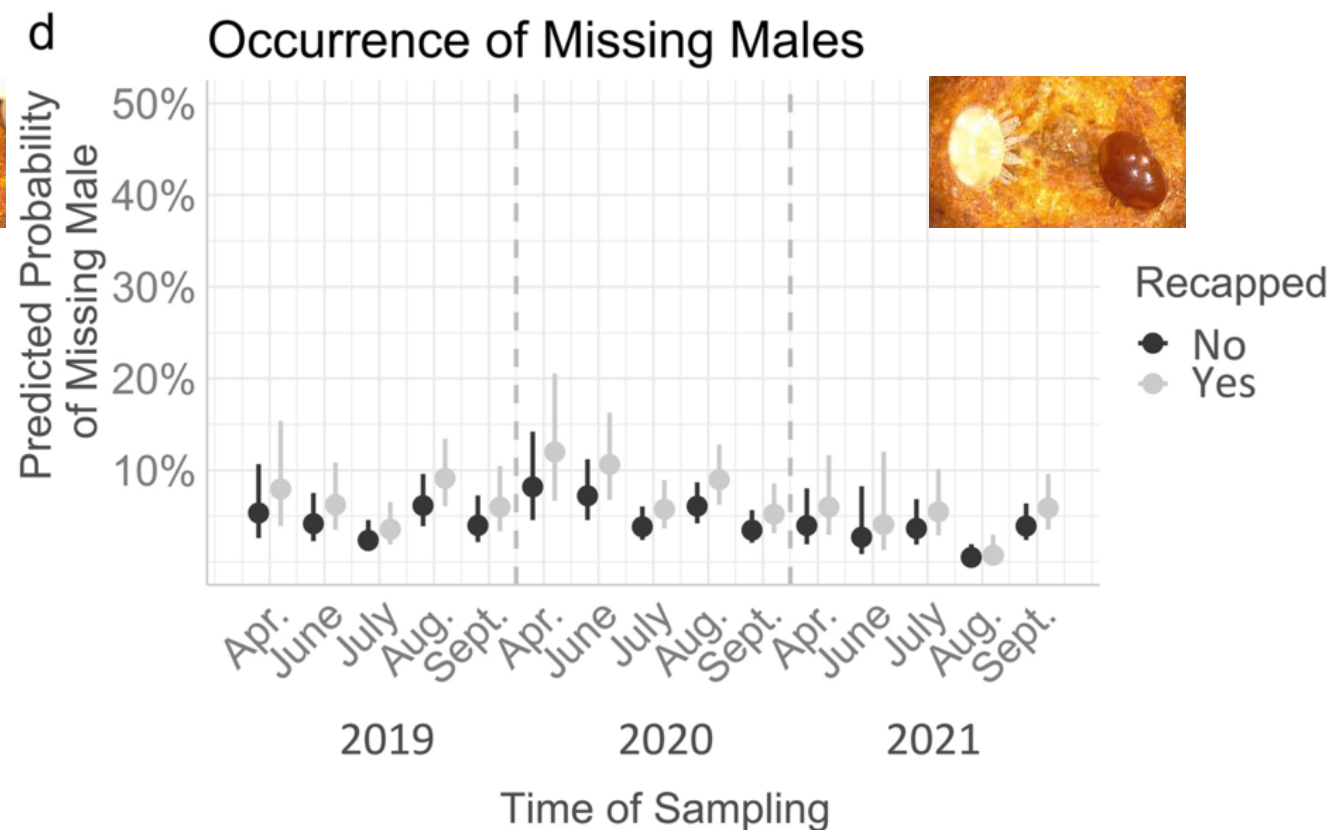
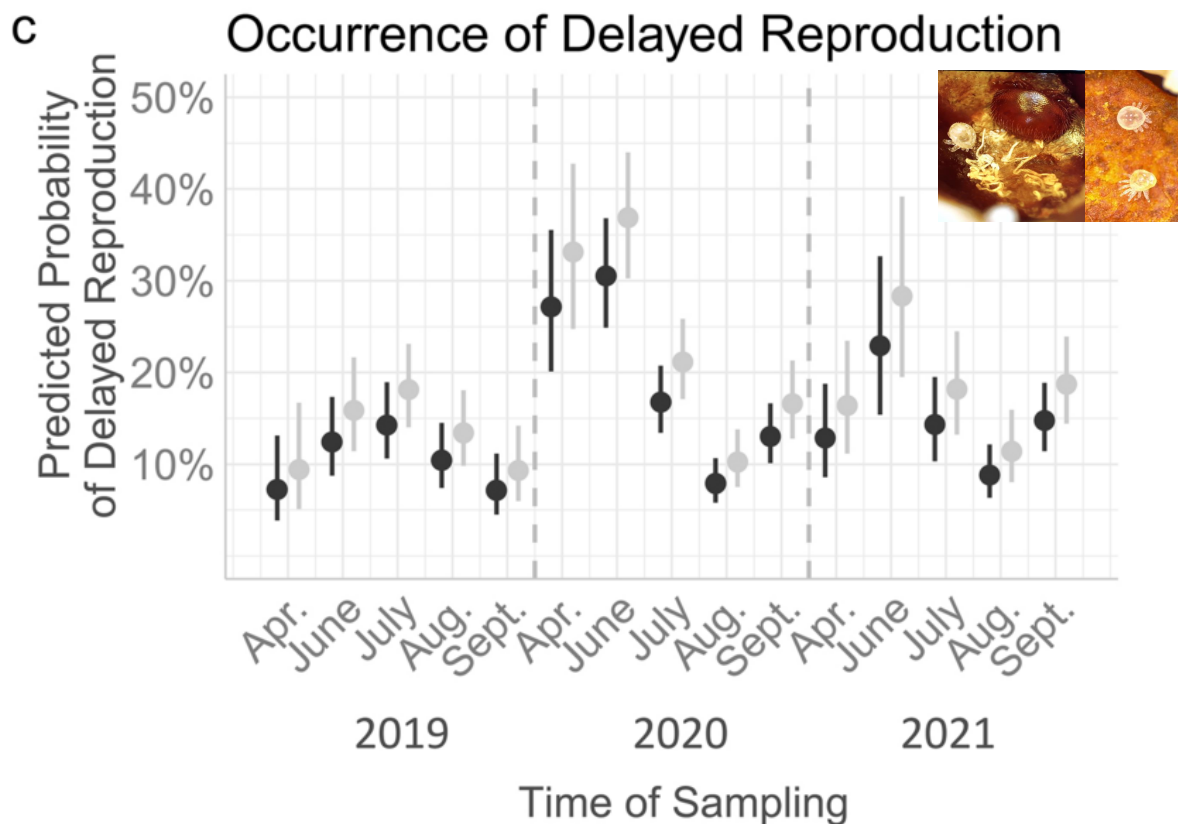
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50	 Old Larva	 Pupa	 Pupa	 Pupa	 REC	 Pupa	 Pupa
52	 Old Larva	 Pupa	 Pupa	 Empty	 Empty	 Nectar	 Nectar



Delayed reproduction and the absence of males are more common in recapped cells



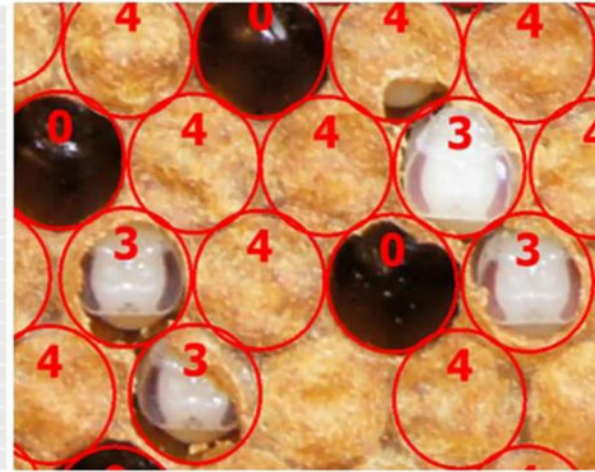
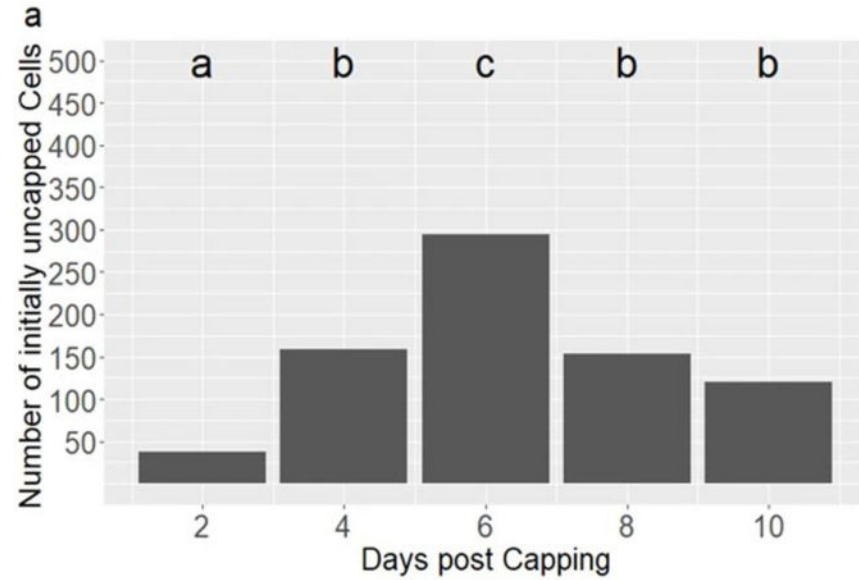
Day 10



GLMM:
Month: $\chi^2 = 120.91$, $df = 14$, $p < 0.001$;
Recapping status: $\chi^2 = 9.15$, $df = 1$, $p = 0.003$; 45 colonies, 95 % CI displayed

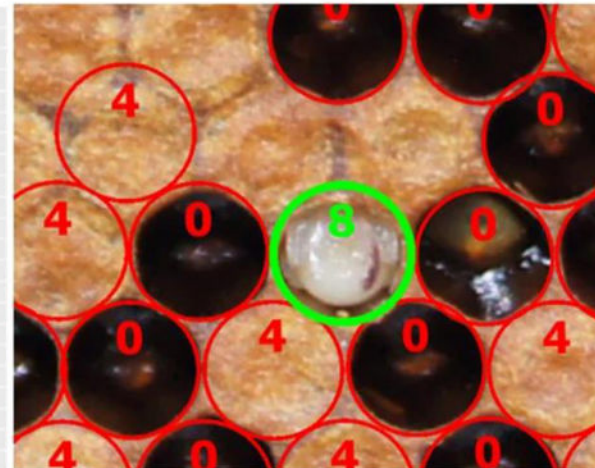
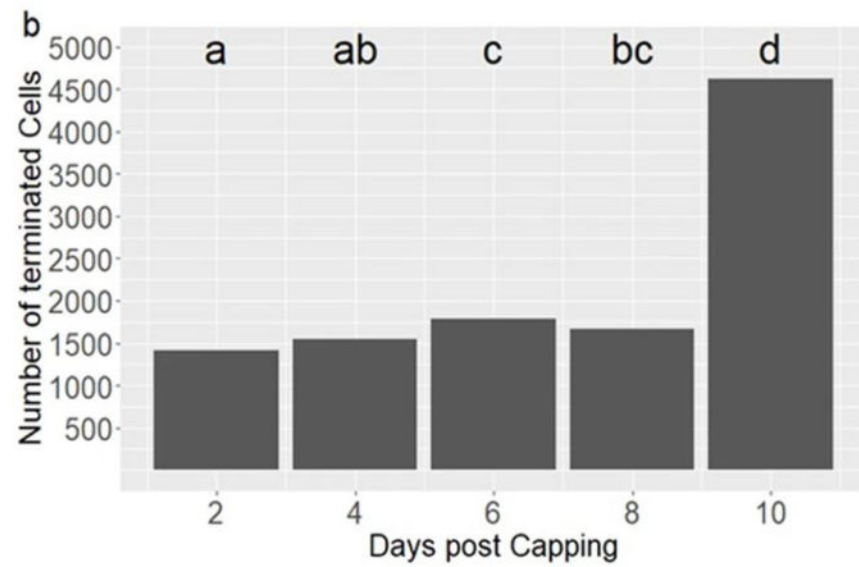
GLMM:
Month: $\chi^2 = 32.22$, $df = 14$, $p = 0.004$;
Recapping status: $\chi^2 = 8.1$, $df = 1$, $p = 0.004$; 45 colonies, 95 % CI displayed

Frequency of recapping and brood termination differs between brood ages



GLMM:

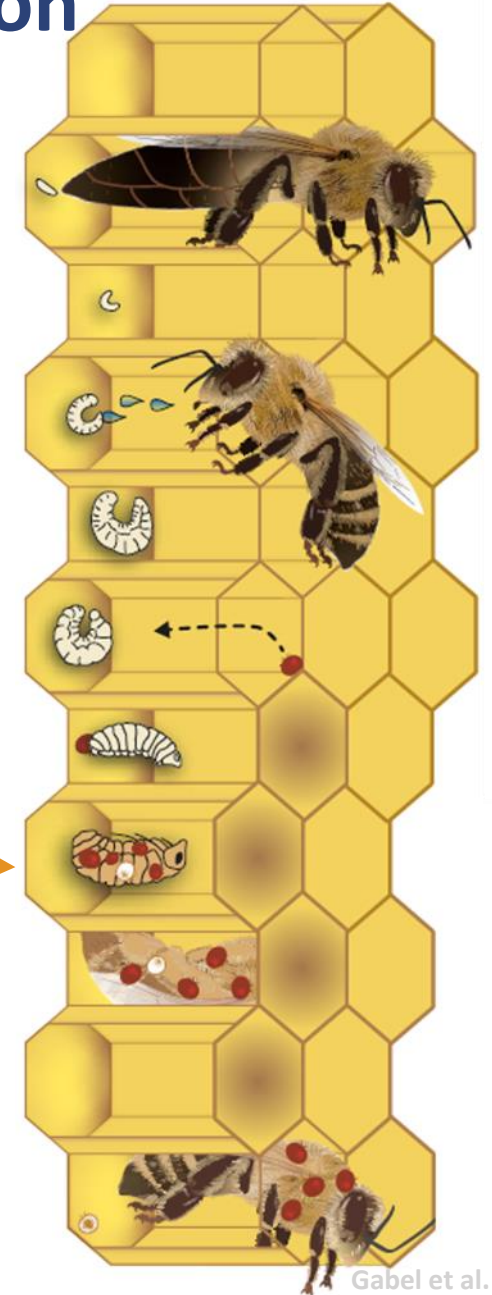
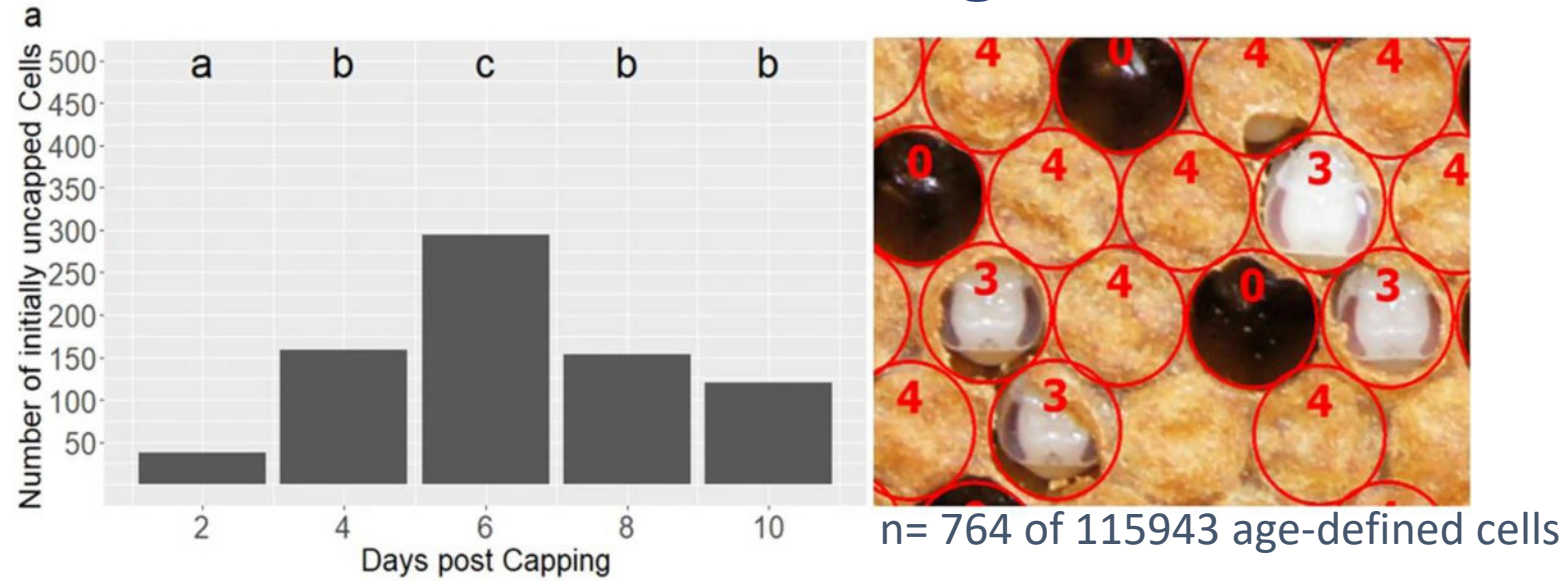
Brood age: $\chi^2 = 238.13$, $df = 4$, $p < 0.001$,
45 colonies, total numbers displayed,
 $n = 764$ of 115943 age-defined cells,
different letters indicate significant
differences between brood ages (Tukey-
Method adjusted for comparing 5
estimates, $p < 0.05$ each)



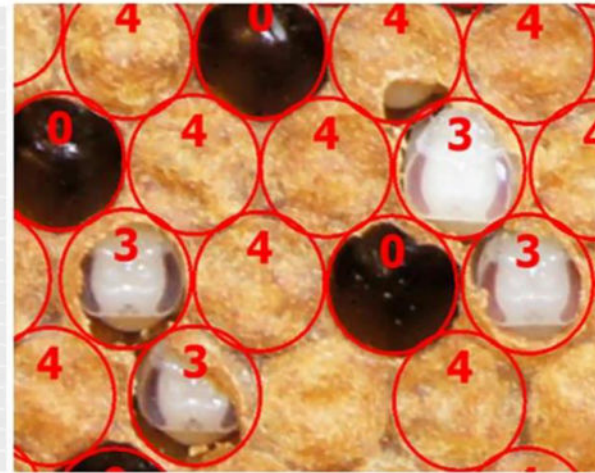
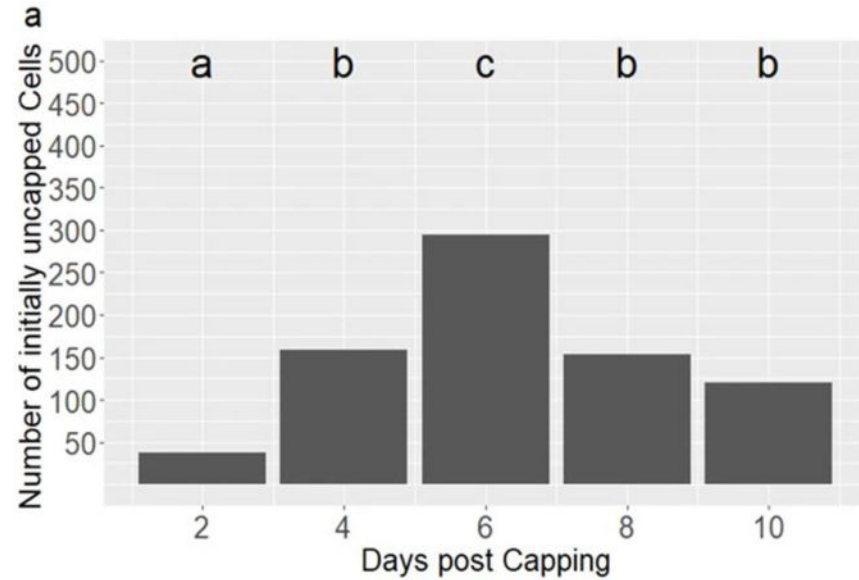
GLMM:

Brood age: $\chi^2 = 3783.6$, $df = 4$, $p < 0.001$,
45 colonies, total numbers displayed,
 $n = 11045$ of 115943 age-defined cells,
different letters indicate significant
differences between brood ages (Tukey-
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Frequency of recapping and brood termination differs between brood ages



Frequency of recapping and brood termination differs between brood ages



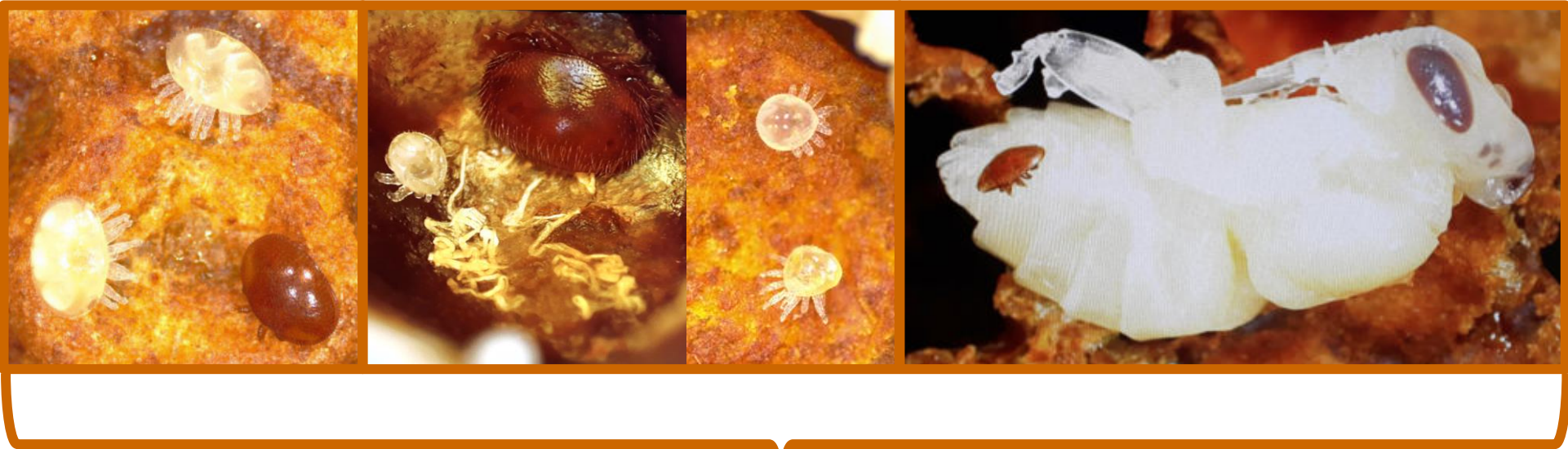
n= 764 of 115943 age-defined cells

Reproductive state	Recapping Frequency of the respective reproductive state	n
Reproductive	40.8 %	2626
Non-reproductive	45.2 %	1480
Delayed	47.5 %	629
Infertile	40.1 %	638
No male	53.5 %	213



Factors denoted in bold indicate significant differences between groups ($p < 0.05$; Tukey-Method adjusted for comparing 4 estimates averaged over sampling time).

Comparison	Estimate	Z	p
Non-reproductive - reproductive	-0.265	-3.510	< 0.001
Infertile - reproductive	0.104	1.000	0.75
Delayed - reproductive	0.338	3.322	0.005
No male - reproductive	0.482	3.073	0.011
Infertile - no male	-0.378	-2.133	0.143
Delayed - infertile	0.234	1.808	0.27
Delayed - no male	-0.144	-0.823	0.844



General Aims:

- Identify mechanisms that lead to decreased mite reproduction
- Investigate effects of recapping behaviour on cell level
- Draw implications for resistance breeding and beekeeping practice

MNR

mite non-reproduction

**other
factors**

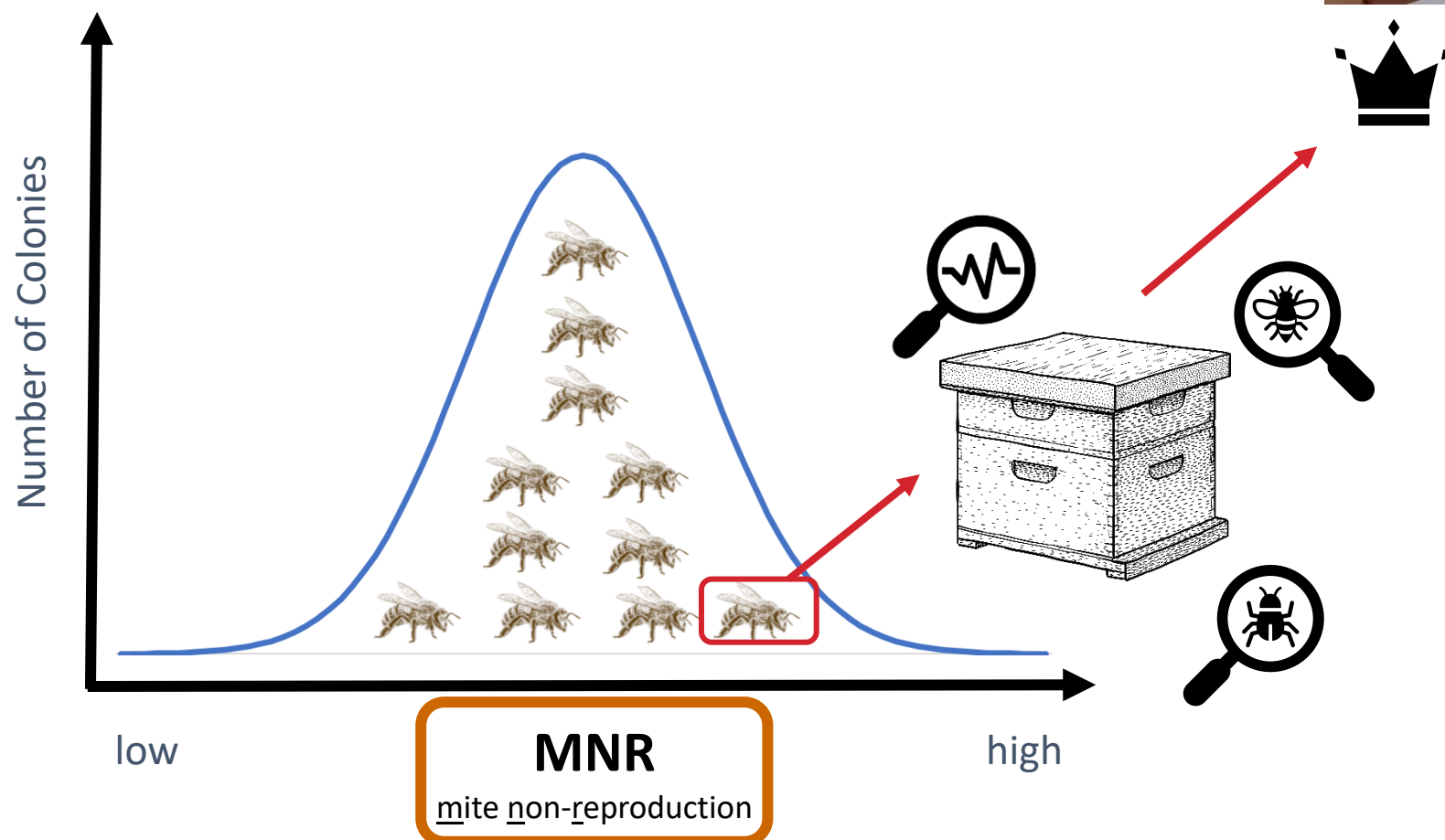


REC

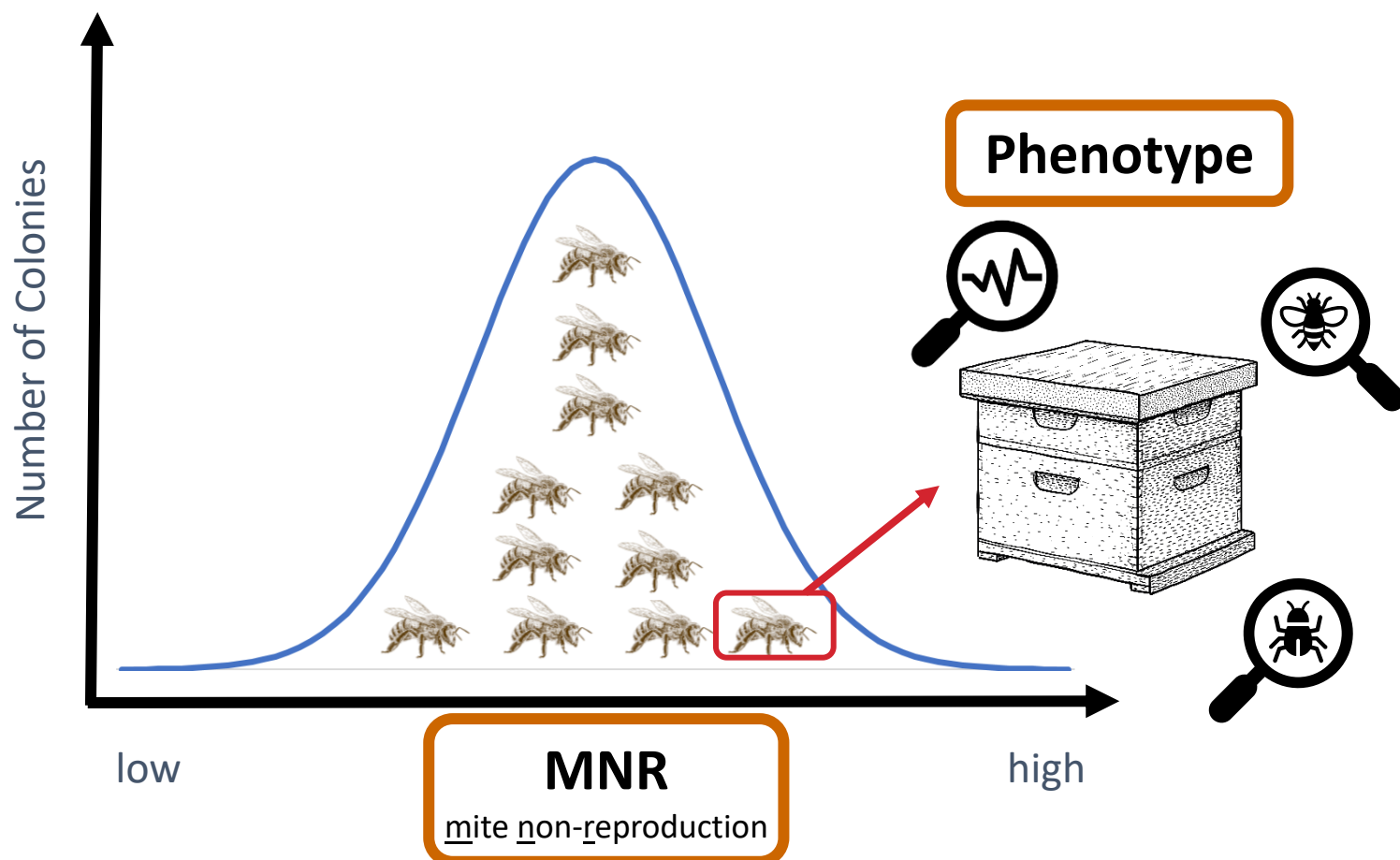
uncapping & recapping
of broodcells



Principles of honey bee breeding



Principles of honey bee breeding



other
factors



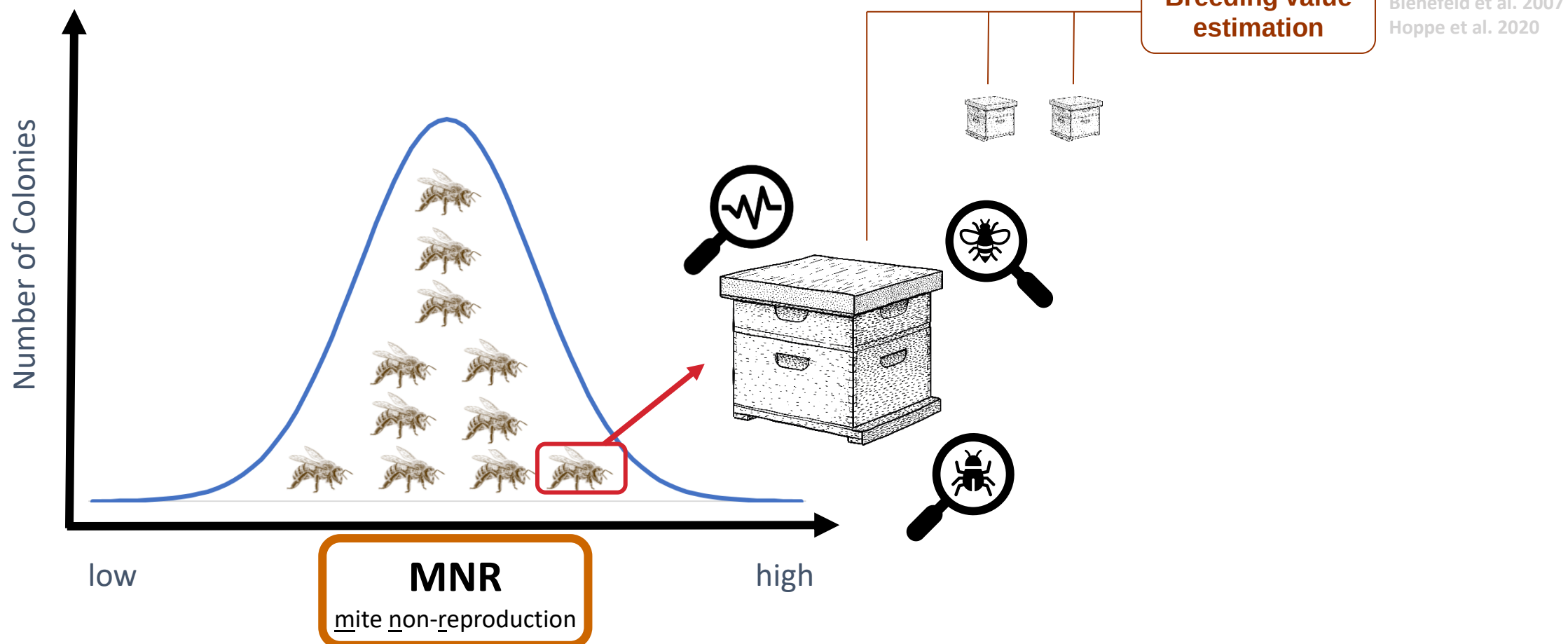
Genotype



Heritability:

The proportion of observed phenotypic variance for which differences in the genotype are responsible (Lush 1945)

Principles of honey bee breeding





Mite non-reproduction and recapping in resistance breeding



Extensive dataset of pedigree and performance test data

- Joint project on MNR-selection in Germany
 - Buckfast and Carnica breeding stocks
 - 1800 tested MiniPlus colonies with single-drone inseminated queens
 - 950 full-grown performance test colonies
-
- Calculation of heritabilities for mite non-reproduction and recapping
 - Implementation of these traits in breeding value estimation



Mite non-reproduction and recapping in resistance breeding

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Heritabilities of MNR & REC appear to be comparable to the heritability of honey yield



Mite non-reproduction and recapping in resistance breeding



Extensive dataset of pedigree and performance test data

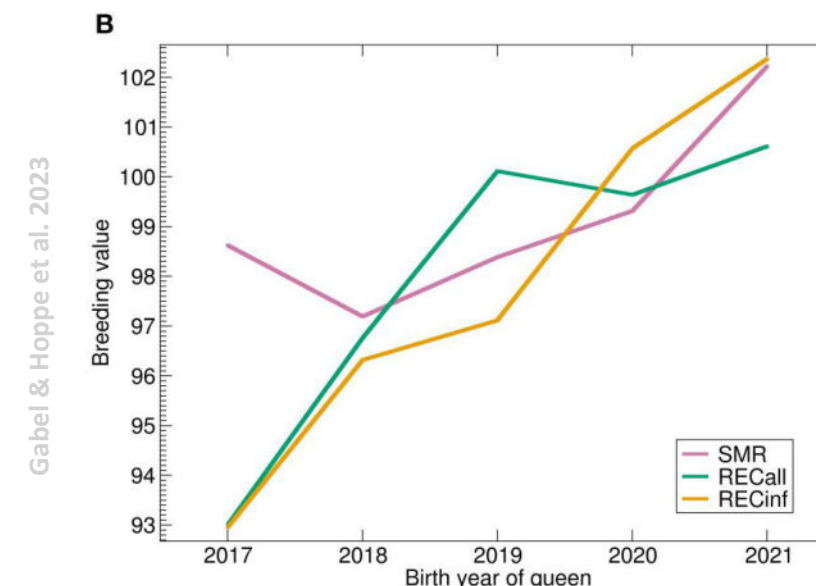
- Joint project on MNR-selection in Germany
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- Calculation of heritabilities for mite non-reproduction and recapping
- Implementation of these traits in breeding value estimation

Breeding Value Estimation enables more accurate selection decisions

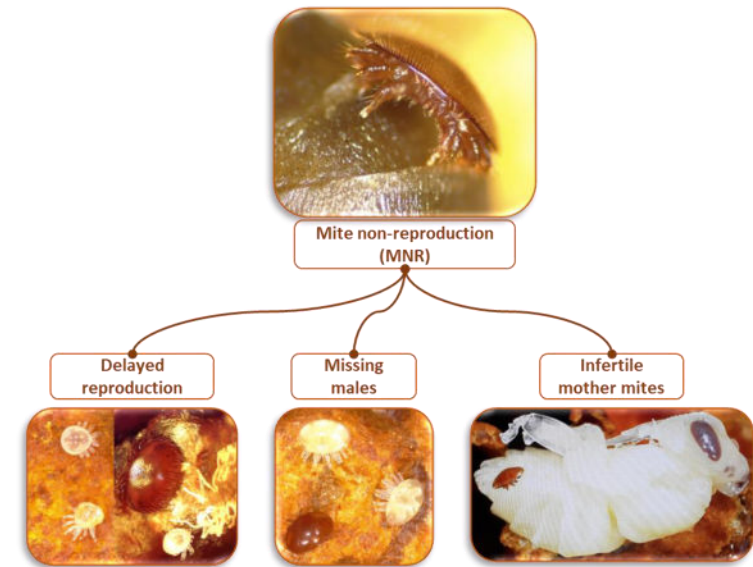


	Breeding value	Reliability	Weighting in %		Breeding value	Reliability	Weighting in %
Honey yield	106	0.50	15	Chalkbrood	105	0.18	
Defensive behavior	119	0.60	15	Chronic Bee Paralysis	101	0.28	
Steadiness on comb	118	0.59	15	Nosemosis	99	0.08	
Swarming drive	106	0.54	15	SMR	115	0.35	
Varroa	115	0.56	40	Recapping	89	0.48	
Total breeding value	117		--				
Performance index	114	0.50					
rate of opened cells	111	0.56					
Varroa infestation development	117	0.46					



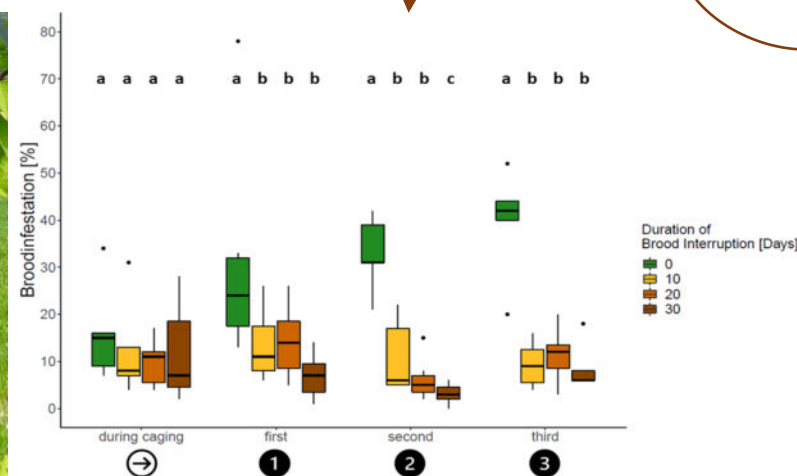
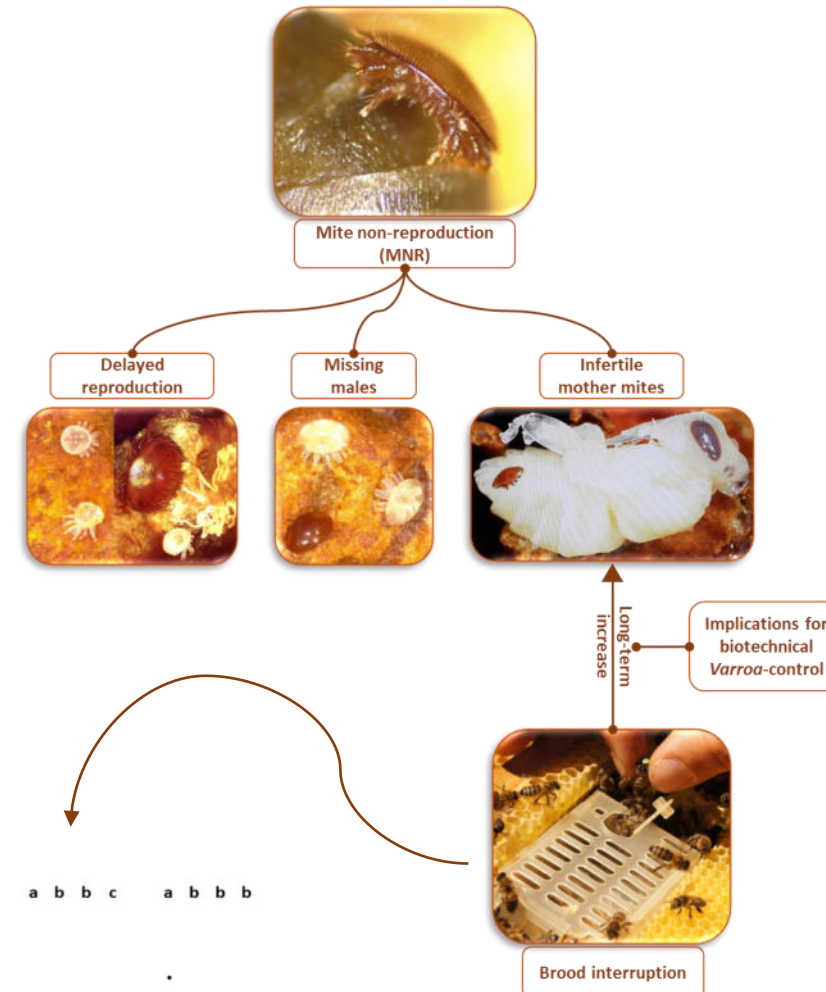
Summary and Conclusion

- Underlying causes of decreased mite reproduction are affected by different factors



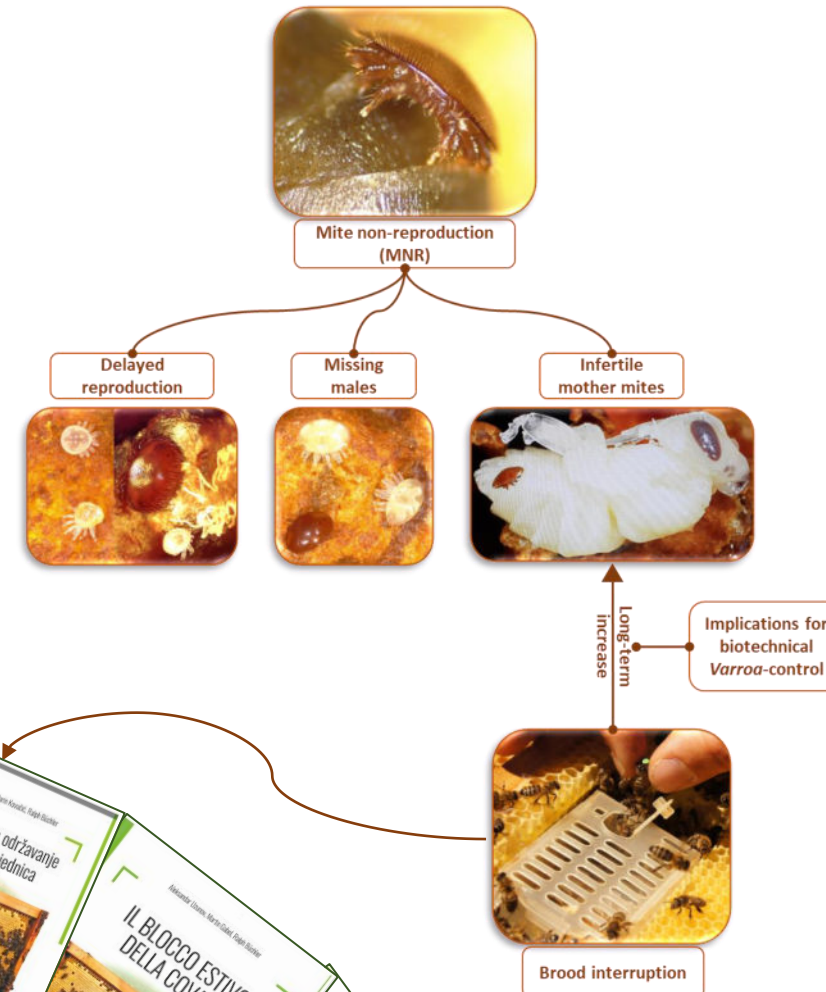
Summary and Conclusion

- Underlying causes of decreased mite reproduction are affected by different factors
 - Brood interruptions mainly increase the occurrence of infertile mother mites



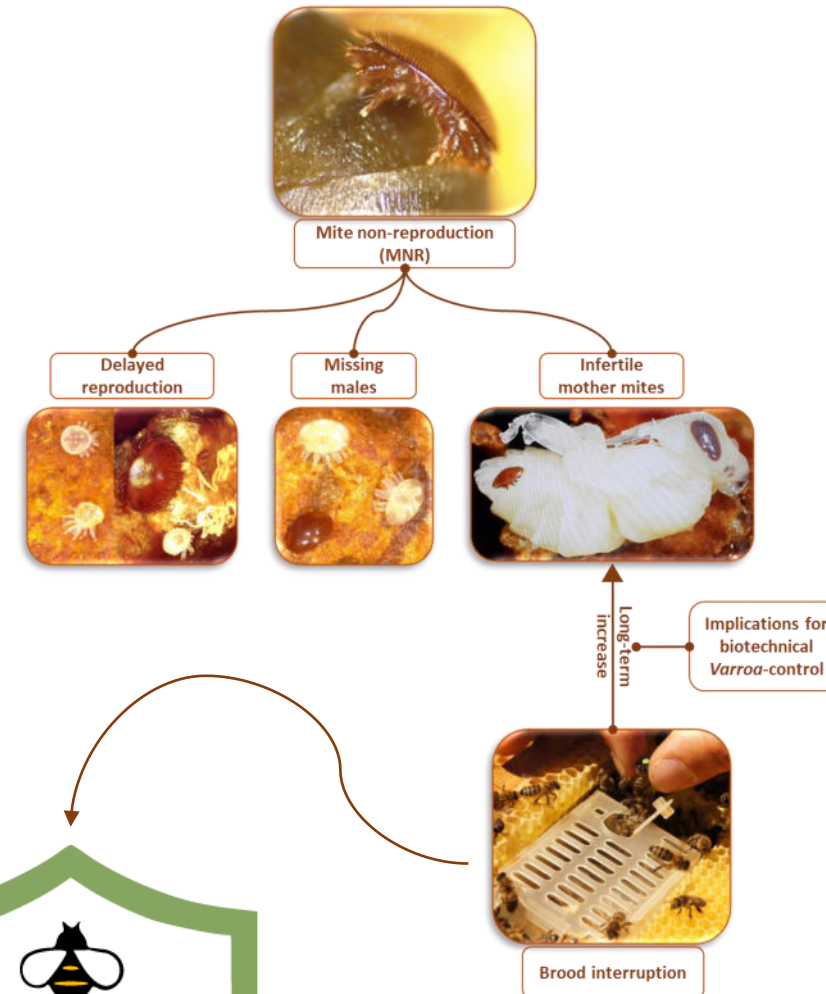
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Summary and Conclusion

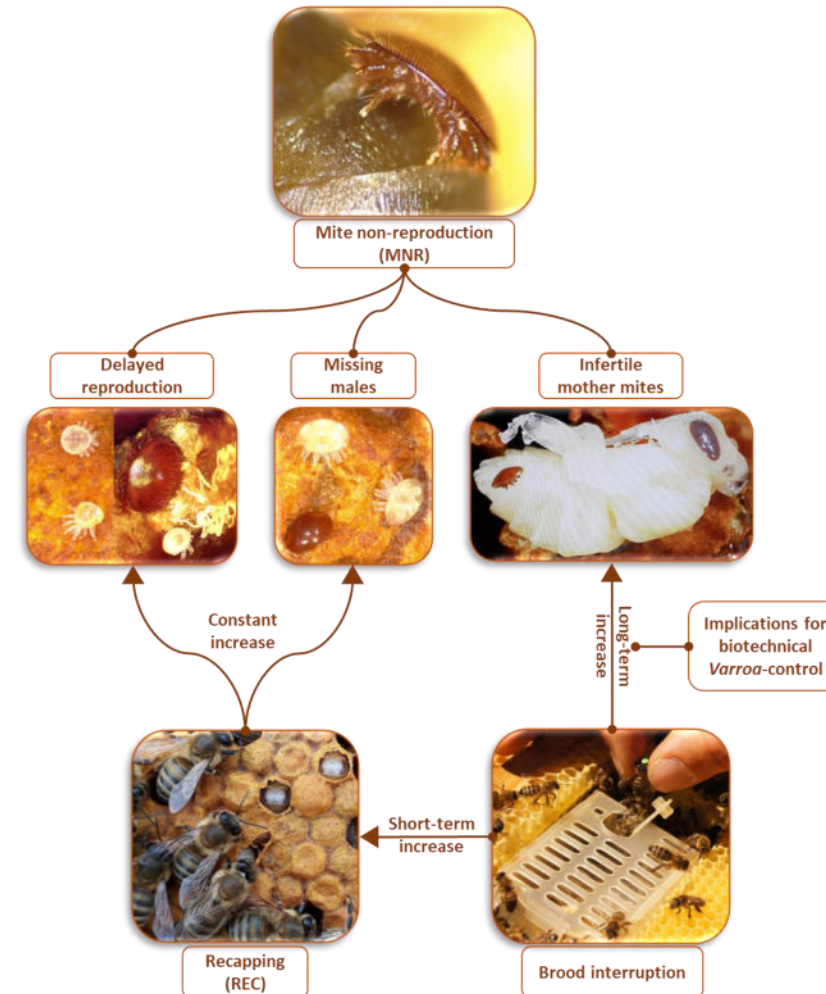
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BEE-GUARDS

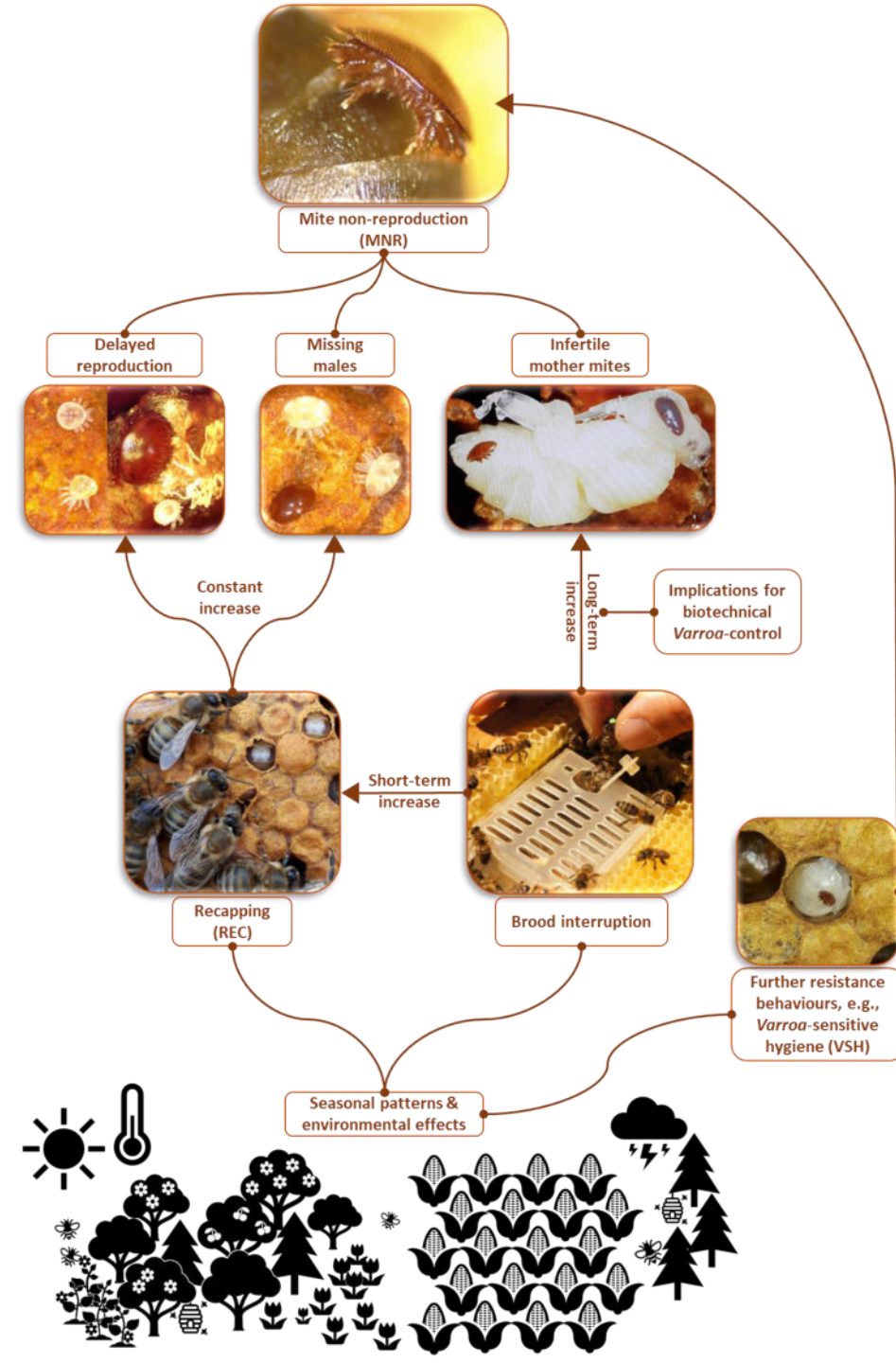
Summary and Conclusion

- Underlying causes of decreased mite reproduction are affected by different factors
 - Brood interruptions mainly increase the occurrence of infertile mothers
 - Recapping mainly affects young stages of mite offspring



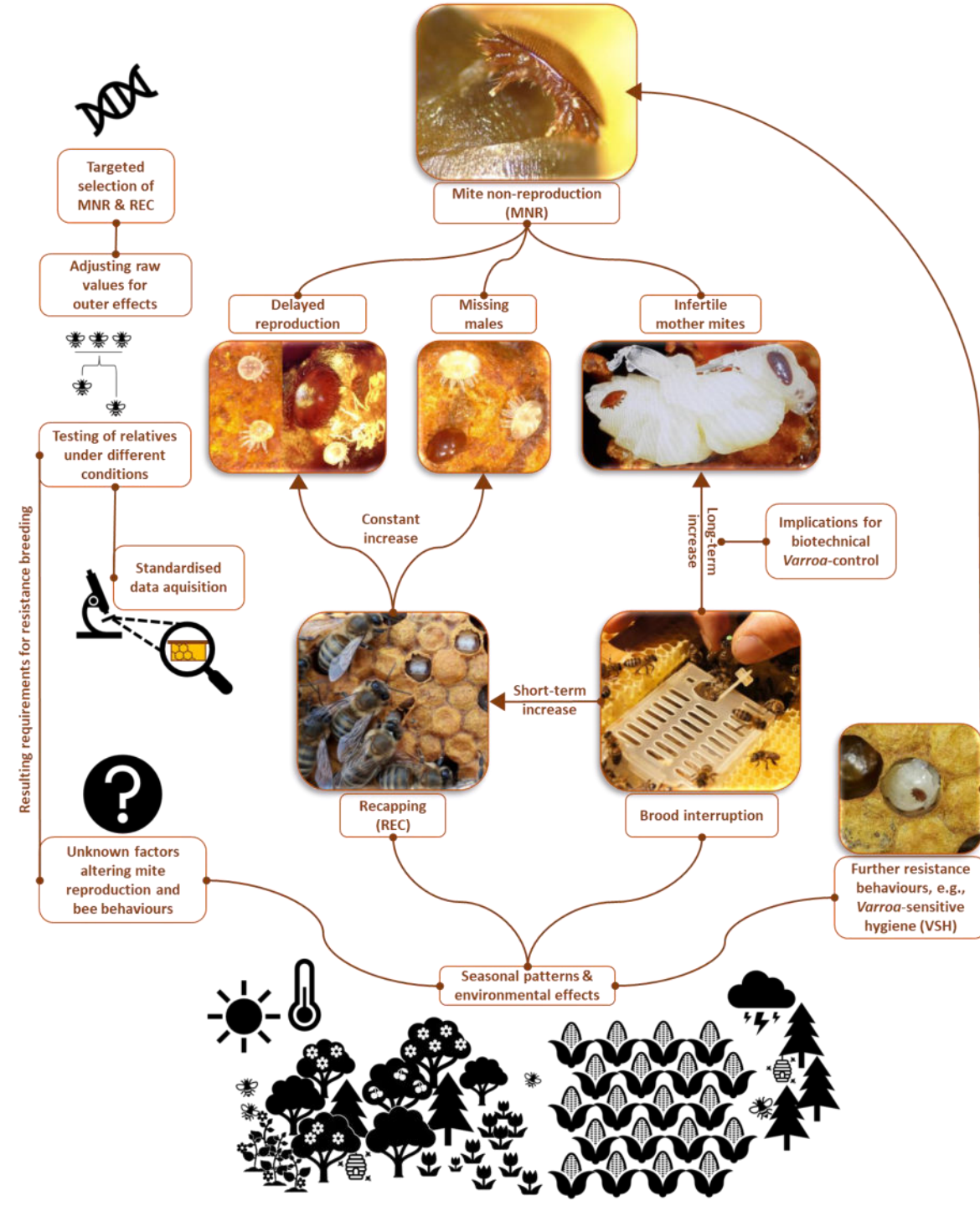
Summary and Conclusion

- Underlying causes of decreased mite reproduction are affected by different factors
 - Brood interruptions mainly increase the occurrence of infertile mothers
 - Recapping mainly affects young stages of mite offspring
- Additional resistance behaviours might alter reproductive success of mites
- MNR and REC are strongly affected by outer effects and display correspondingly low heritabilities



Summary and Conclusion

- Underlying causes of decreased mite reproduction are affected by different factors
 - Brood interruptions mainly increase the occurrence of infertile mother mites
 - Recapping mainly affects young stages of mite offspring
- Additional resistance behaviours might alter reproductive success of mites
- MNR and REC are strongly affected by outer effects and display correspondingly low heritabilities
- Targeted selection of MNR and REC thus relies on large-scale testing and breeding value estimation





Many
Thanks to:

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Julius-Maximilians-
**UNIVERSITÄT
WÜRZBURG**

With support from

 Federal Ministry
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Questions
?



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