Beekeeping with Varroa in the Netherlands: Resilience, Rewilding and Conservation

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Wageningen University and Research, The Netherlands

Scottish Native Honey Bee Society, Winter meeting, 21 February 2022



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Inbuzz
Imkersbedrijf

Beekeeping in the Netherlands

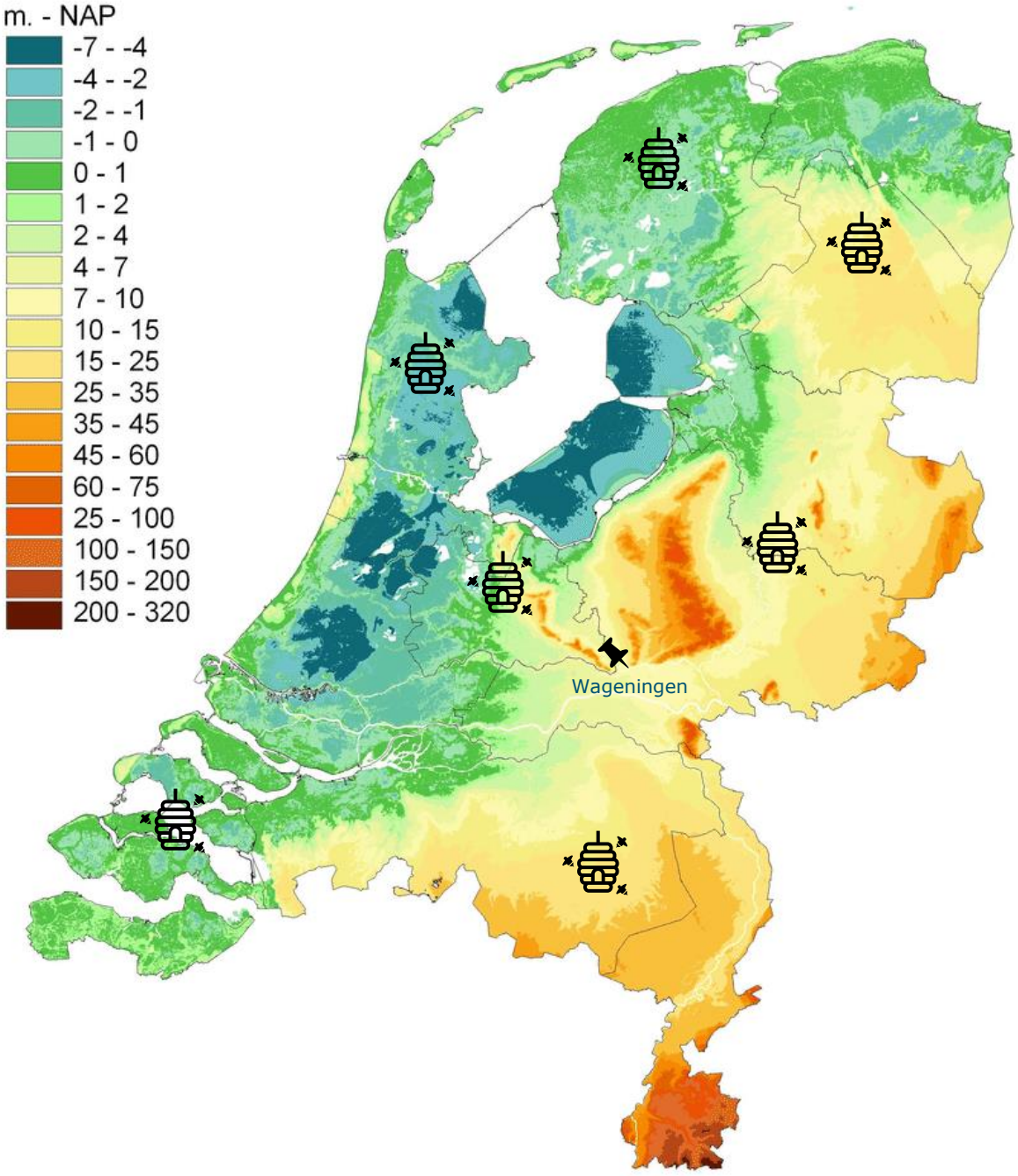
- ≈ 80 000 managed colonies
- ≈10 800 beekeepers
- ≤20 professional operations

- ≈ 200 local beekeepers' associations



Veneendaal bee market, end of 19th century

- 200 000 colonies in 1877!

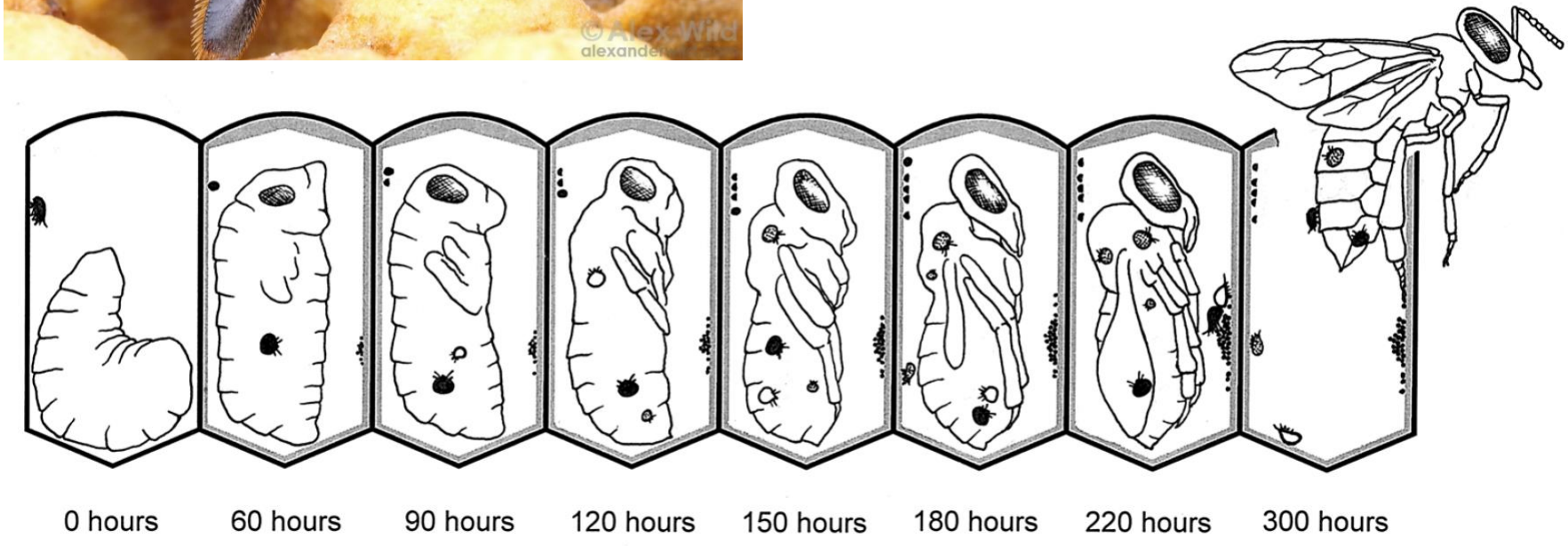


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Varroa destructor







Ben Oldroyd, 1999

Ramsey *et al.*, 2019

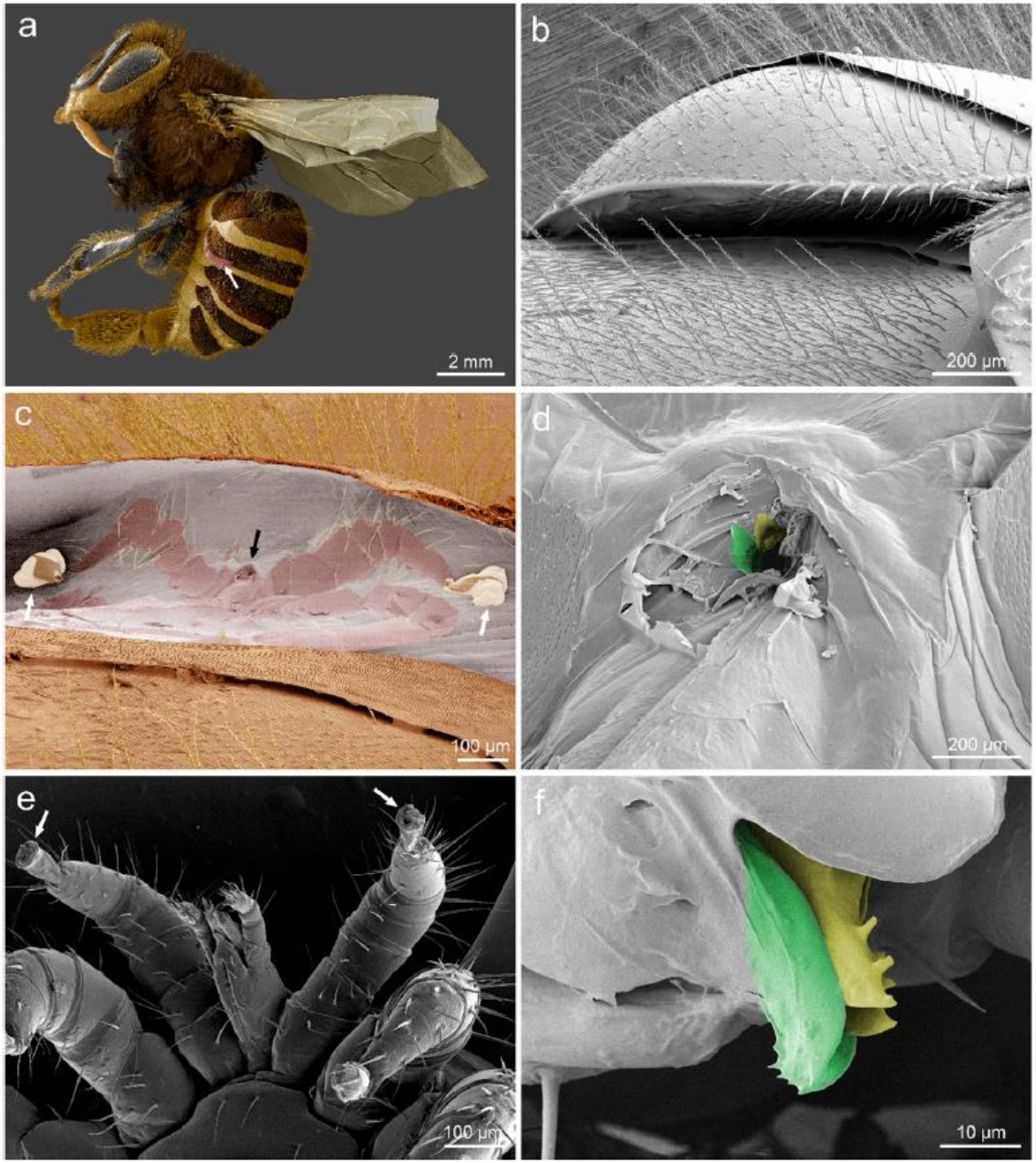
A et B: Mite location on the honeybee (beneath the 3rd tergite)

C: when removed, clear marks of the mite, wound where mouthparts were and attached ambulacra (‘foot pads’)

D: Open wound with Varroa mouthparts (yellow and green)

E: Ambulacra (‘foot pads’)

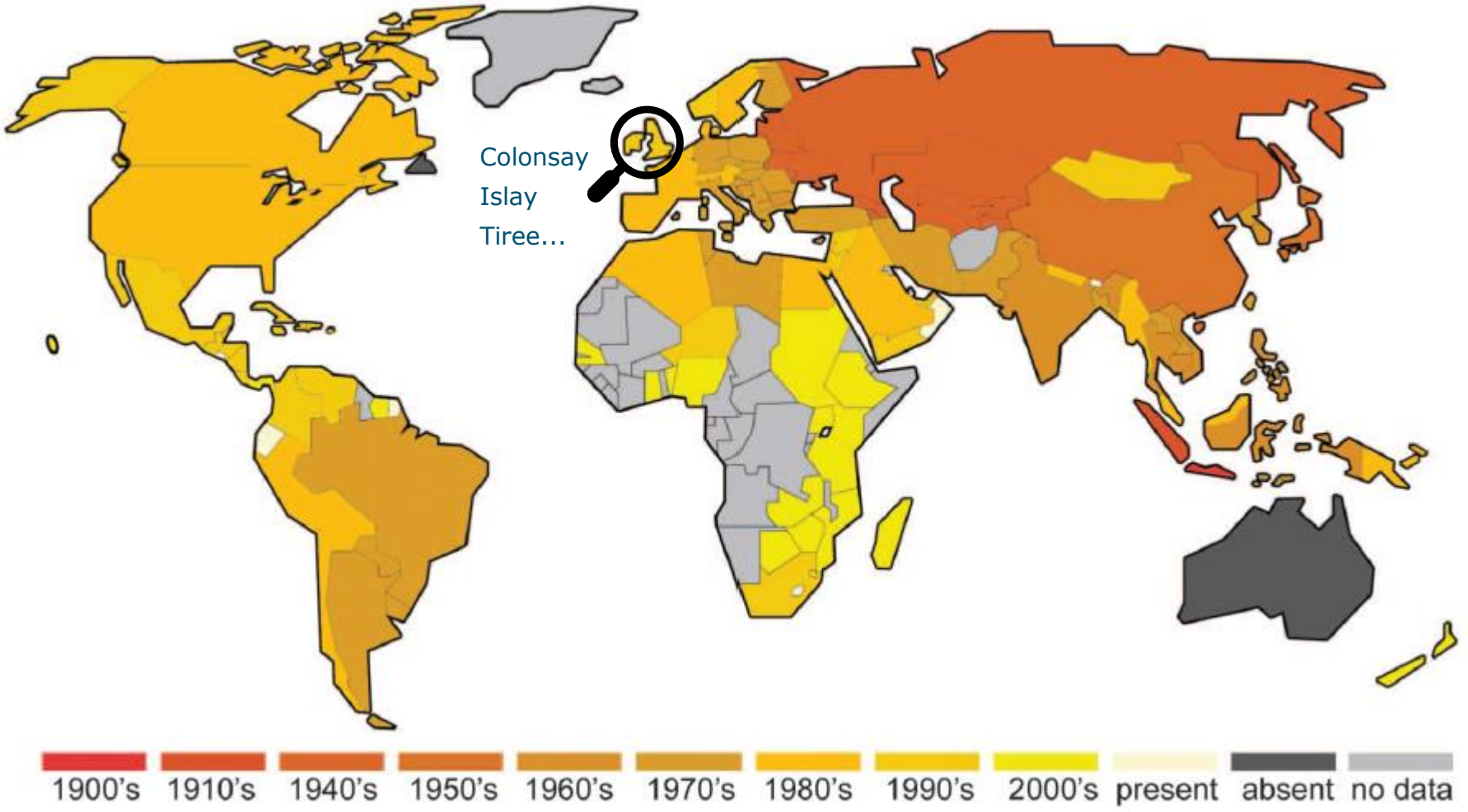
F: Chelicera (mouth parts)



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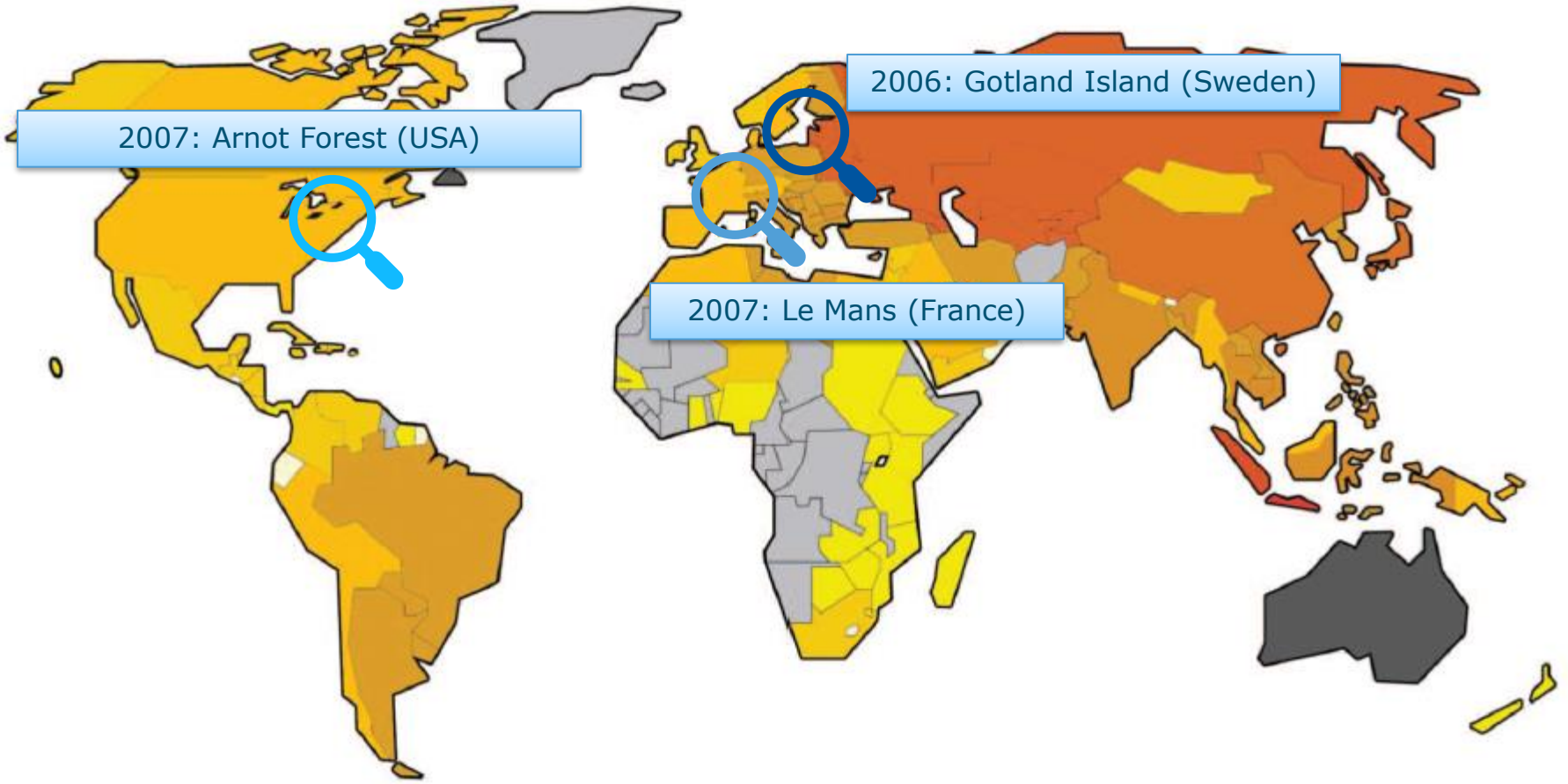
Global spread of *Varroa destructor*

Well, not entirely... Some small islands still hold out against the invader



Wilfert *et al.*, 2016

Global spread of *Varroa destructor*



In the 2000's, reports of survivors start to appear

Sweden: Gotland island



- 150 colonies, 'live and let die' experiment
- Varroa since 1999
- 75%mortality on 3rd year

=> Can it be applied in a beekeeping context?

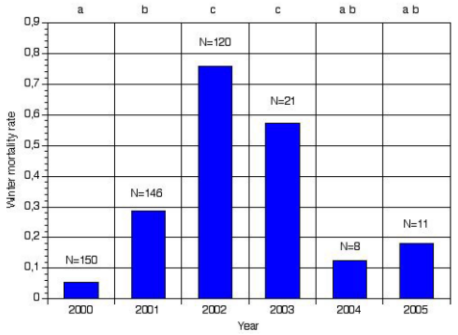


Figure 1. Mortality rate of honey bee colonies over six winters without control of *Varroa destructor*. N = number of colonies in late October each year. Bars with different letters above are significantly different between years ($P < 0.05$, chi-square).

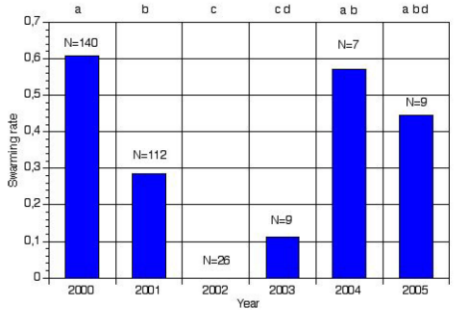


Figure 2. Swarming rate of honey bee colonies for six years without *V. destructor* control. N = number of colonies in late May each year. Bars with different letters above are significantly different between years ($P < 0.05$, chi-square).

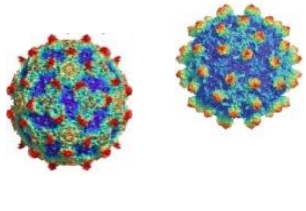


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Why select for resilient colonies?

- Important damage to colonies
  - Direct: feeding behaviour
  - Secondary: Transmission of several viruses
  - Collapse of colonies in 2-3 years







- Need for constant control
  - Risk of rapid resistance to acaricides
  - May prevent host-parasite coevolutionary dynamics

How does it work?



‘Natural’ selection

- ‘passive’ process => no chosen traits
- Approach based on fitness (in evolutionary sense)

Criteria: **Growth** and **reproduction**

- all survivors stay (~50-75%)

≠

Targeted selection

- Active process => chosen traits
- step by step
- only the ~5% ‘best’ stay



Natural selection does not grant organisms what they "need".

Mimicking natural selection

Lelystad population 2007-2022

Conditions:

Complete stop of *Varroa* control

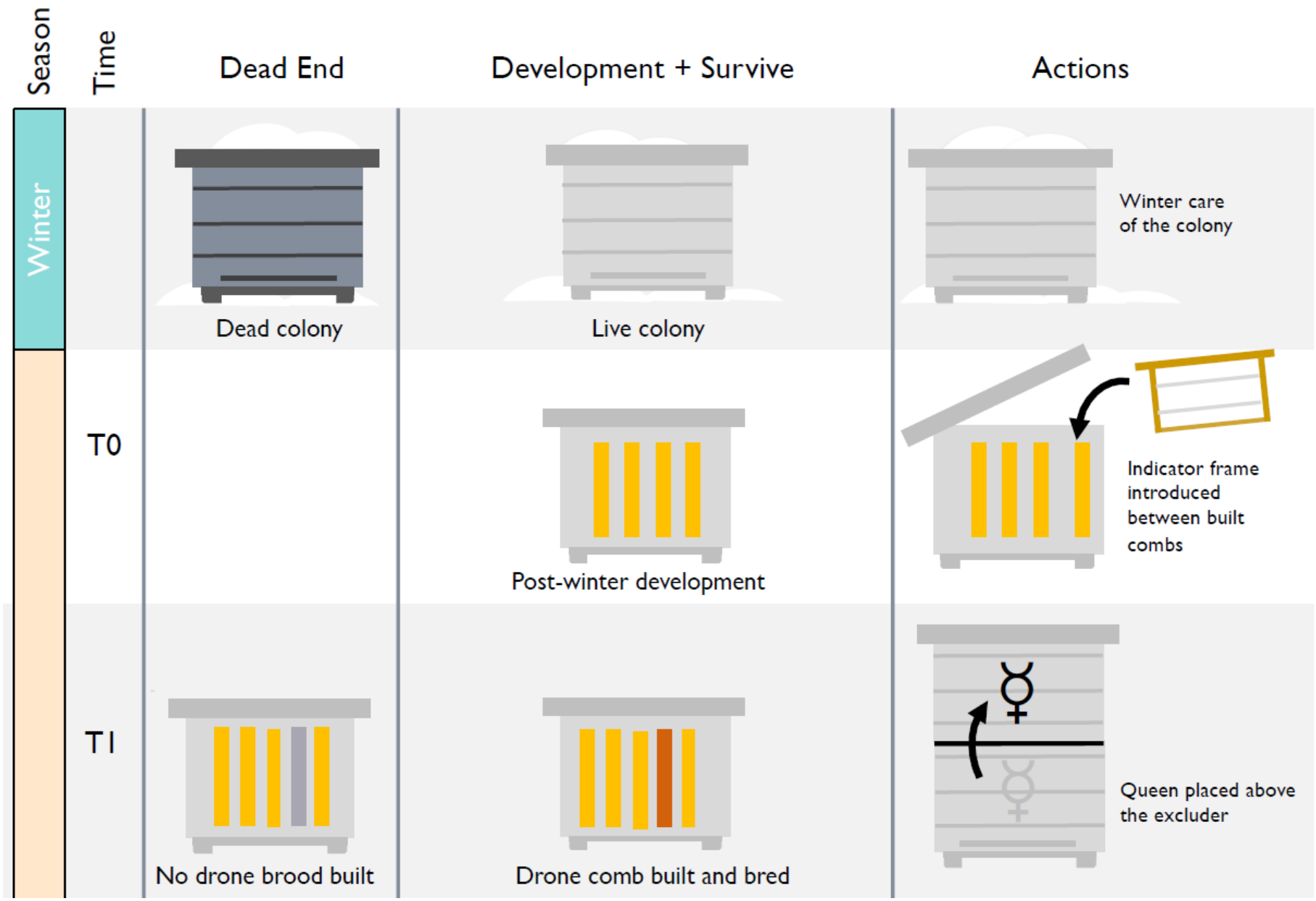
'Selection group' of vital colonies:

- Survival
- good growth
- reproduction (drone comb!)
- splitting and mating (island)

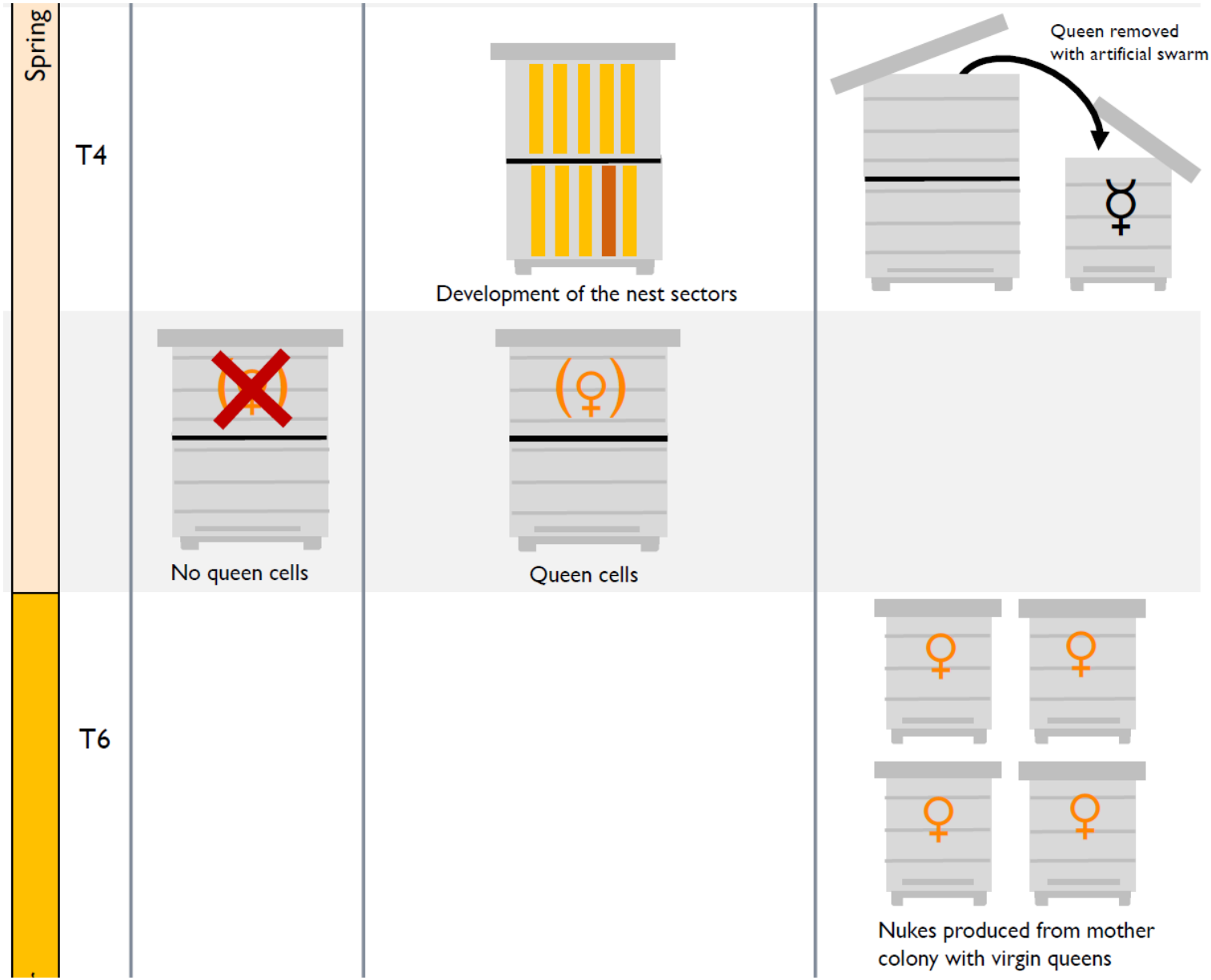
Control:

- Same protocol
- Separate mating area
- Varroa control 2X per year (OA)

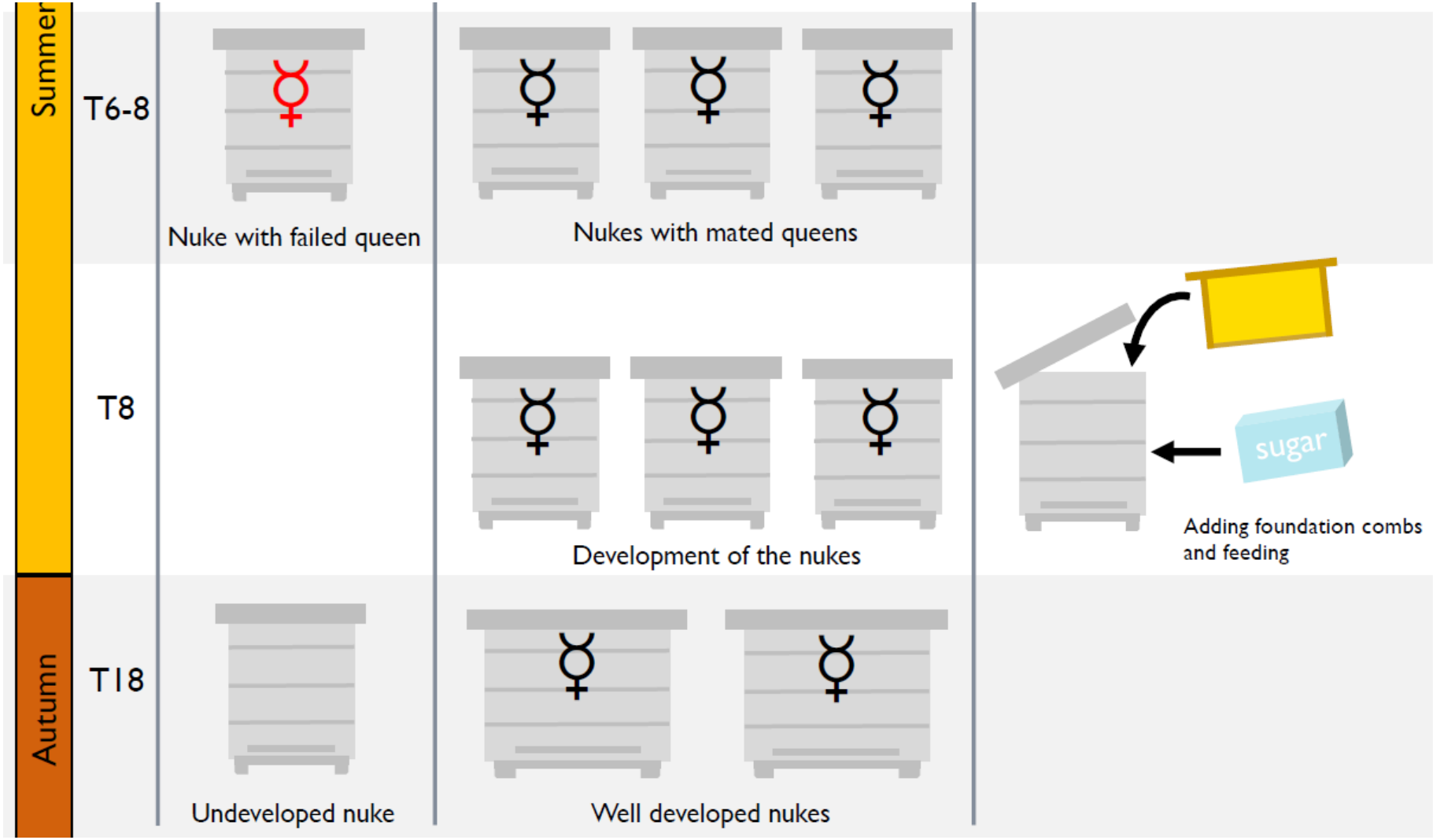




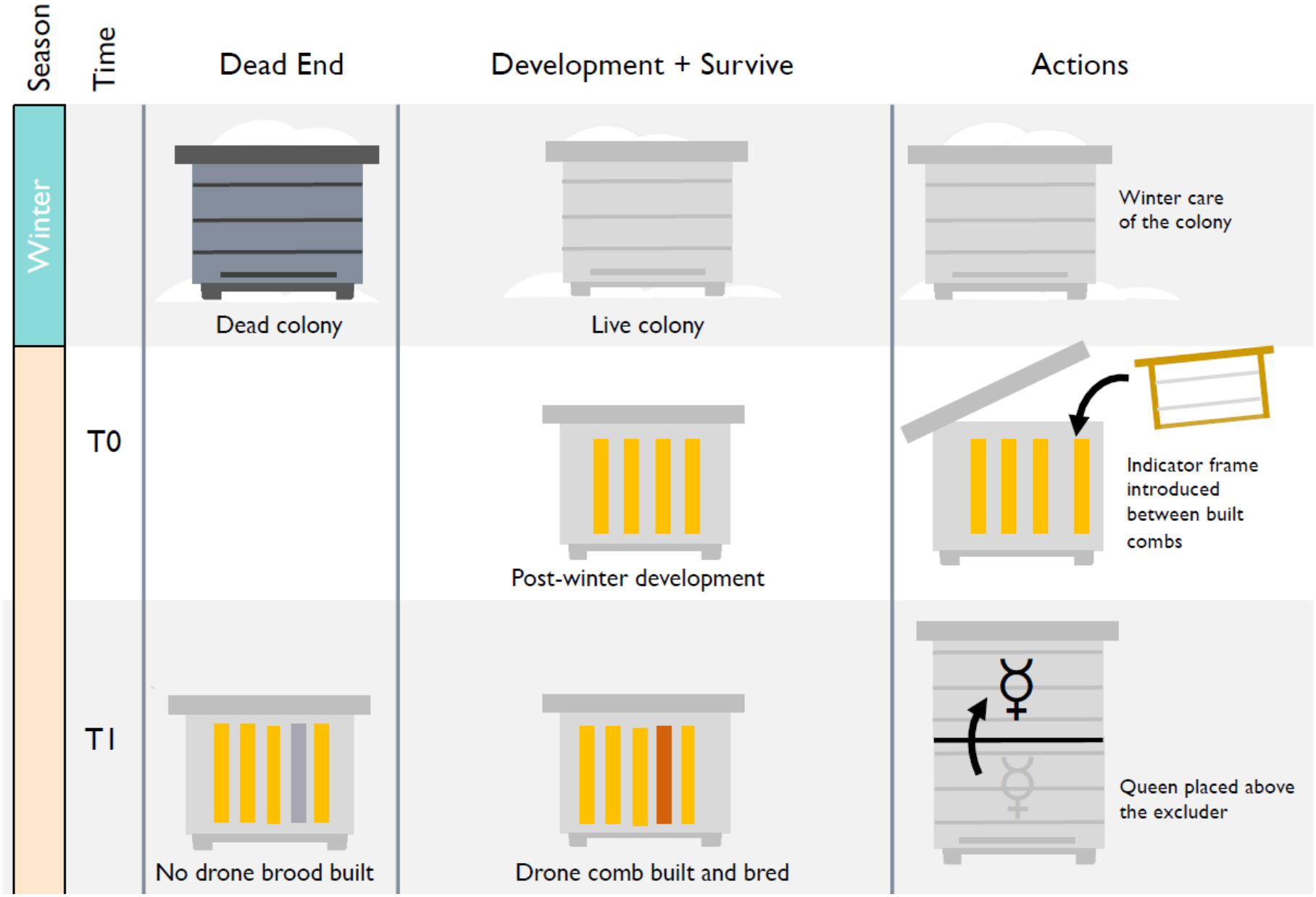
Blacquière *et al.*, (2019) "Darwinian black box selection for resistance to settled invasive Varroa destructor parasites in honey bees." *Biological invasions* 21, no. 8: 2519-2528.



Blacquière *et al.*, (2019) "Darwinian black box selection for resistance to settled invasive Varroa destructor parasites in honey bees." *Biological invasions* 21, no. 8: 2519-2528.



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Blacquière *et al.*, (2019) "Darwinian black box selection for resistance to settled invasive Varroa destructor parasites in honey bees." *Biological invasions* 21, no. 8: 2519-2528.


National Park Hoge Veluwe
(July 2017)



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Take care!

- Find an isolated mating area



- Do not use all your colonies

- Queens from treated apiaries can be replaced gradually

Selected effects

- Colonies survival
    - Growth / survival
    - Reproduction

- Selection against Varroa:
    - **resistance** (number of *Varroa* ↓)
    - **tolerance** (damaged caused by *Varroa* ↓)

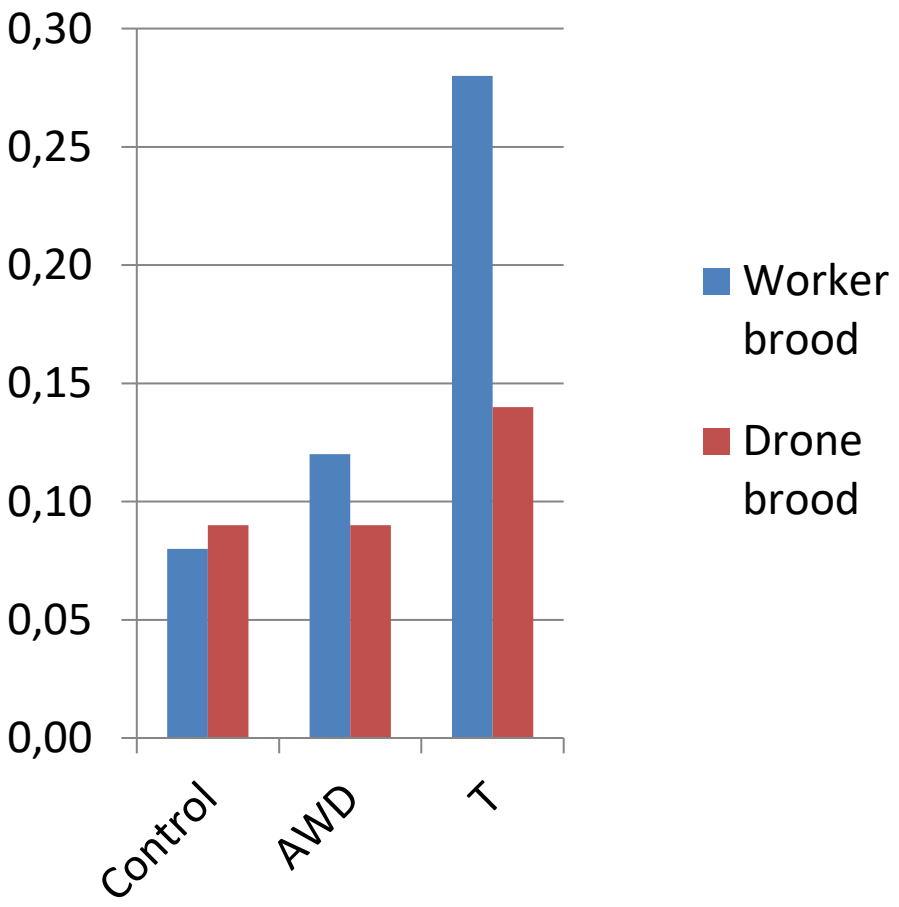
- What are the mechanisms?
- Do they differ between populations?



Reduction of Varroa fertility (non-reproduction)

Proportion of Varroa mites failing to reproduce (worker and drone brood):

	Control	AWD	TG
Worker brood 2011 (Michiel) 2014 (Tjeerd + Chula)	0.03 (N=231) 0.09 (N=109)	0.06 (N=180) 0.15 (N=155)	0.09 (N=409) 0.63 (N=124)
Drone brood 2014 (Tjeerd + Chula)	0.12 (N=496)	0.14 (N=472)	0.21 (N=891)
Worker brood 2012 (Thijs)	0.12 (N=406)	0.16 (N=303)	0.12 (N=417)
Drone brood 2013 (Anne)	0.16 (N=75)	0.04 (N=100)	0.12(N=100)
Drone brood 2015 (Katousha)	0.0 (n=11)	0.10 (N=91)	0.10 (N=91)

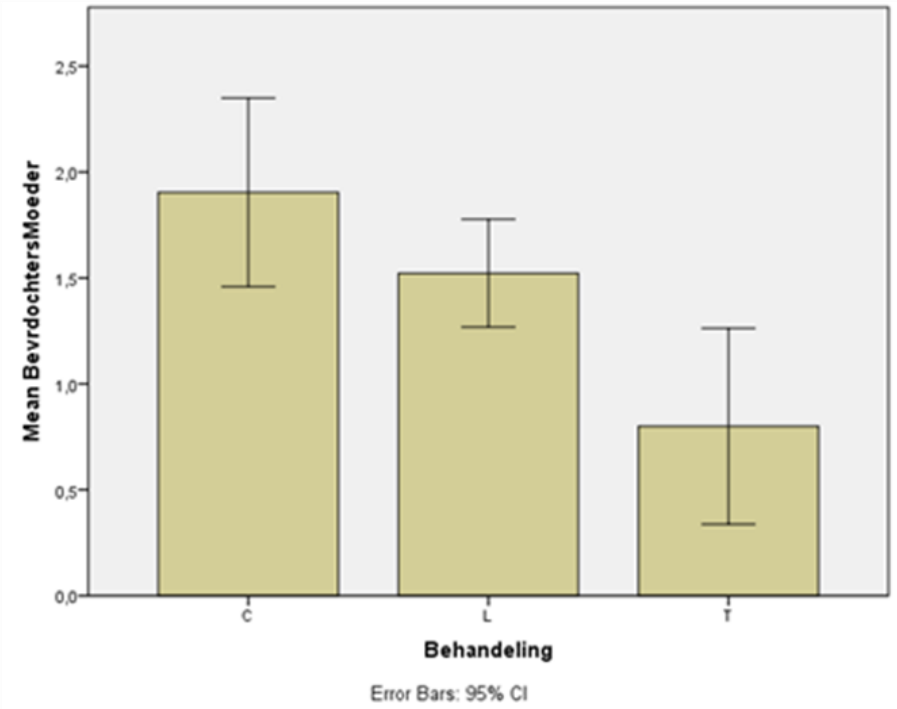


Reduction of fecundity (number of offspring)

	Control	AWD	TG	P
Worker brood 2011 (Michiel)	2.49[a]	1.92[b]	2.02[b]	<0.001
Worker brood 2012 (Thijs)	1.22[b]	1.10[b]	0.97[a]	<0.05

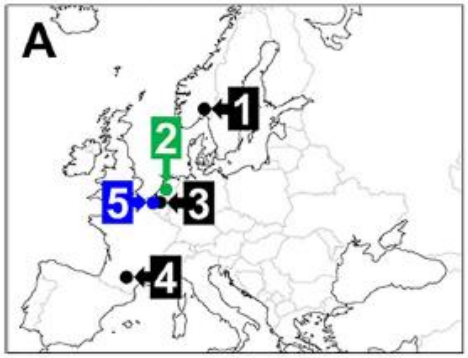


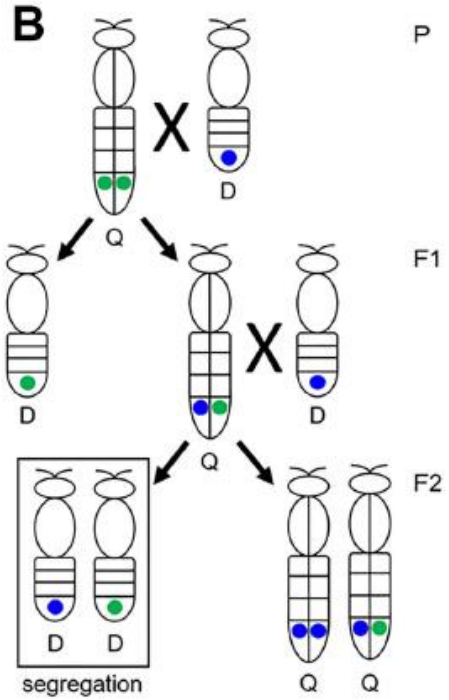
Nombre de descendantes femelles par Varroa dans le couvain d'ouvrières (N = 196, 2014 Tjeerd)



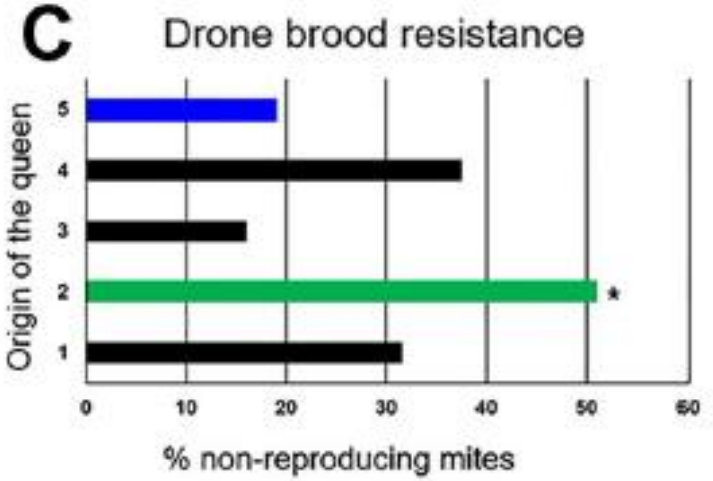
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Non-reproduction in drone brood (Broeckx 2019)





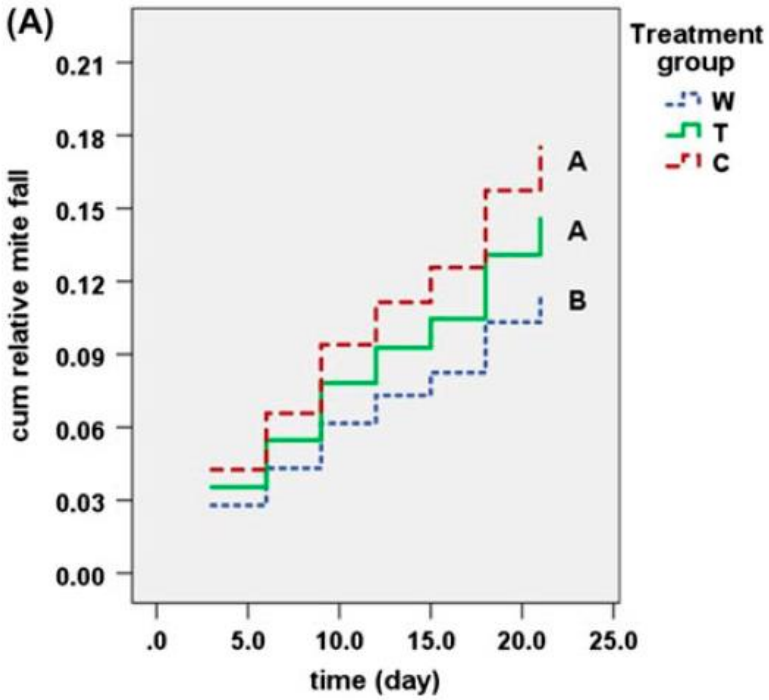
- Trait of drones preventing mite reproduction (chemical signals)

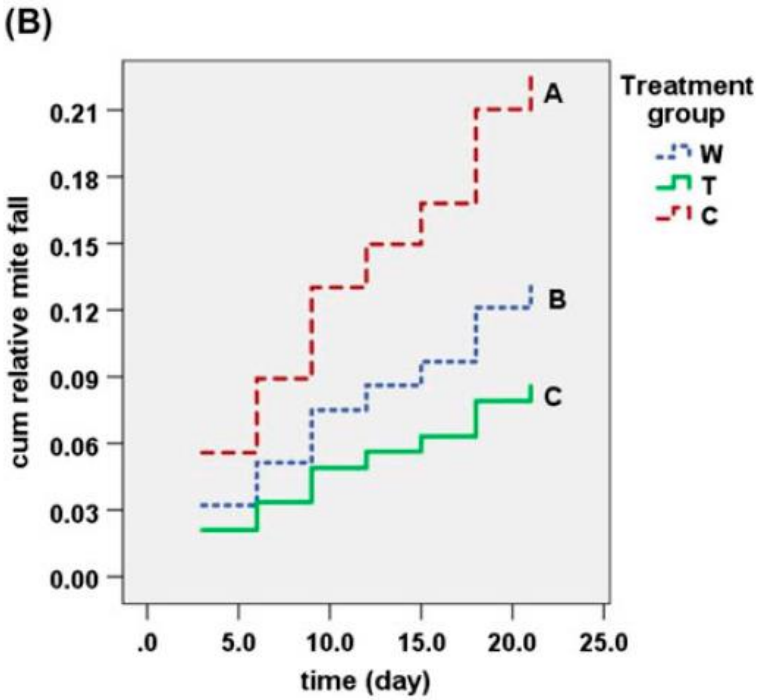


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Grooming (Astrid Kruitwagen, 2015)

- Individual bees: starch experiment
- Bees in groups + *Varroa* (neutral)
- Colonies + phoretic mites





Varroa sensitive hygiene (VSH) (2015)

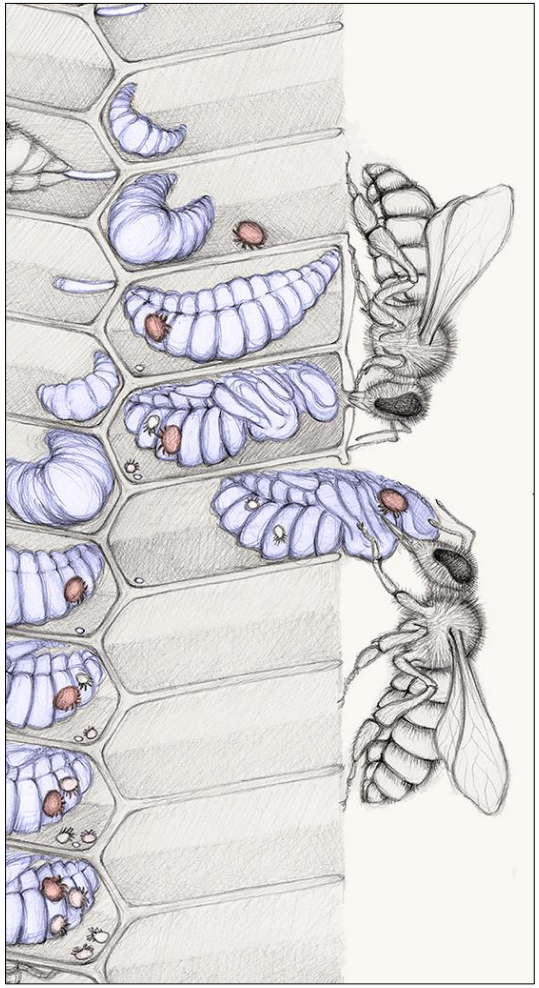


Illustration: Arista bee, NL

- Detection and removal of brood infested by Varroa
- Bees between 6 and 18 days old (average= 11.13 days)
- Seems to be damage-dependent, but the triggers remain unclear

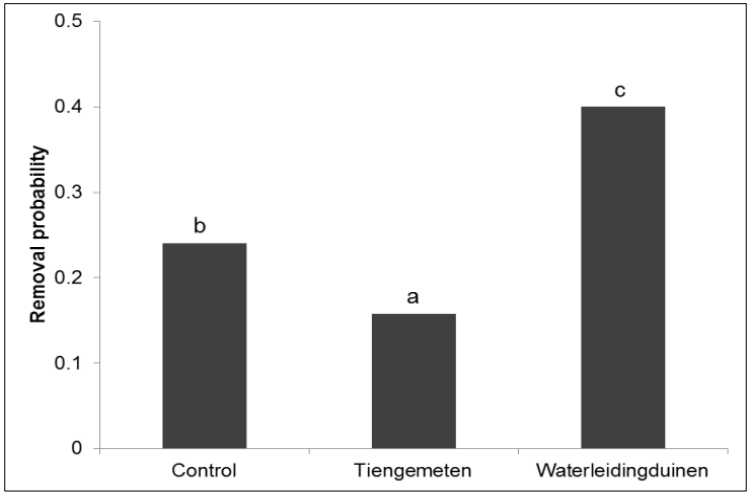
Varroa sensitive hygiene (VSH) (2015)

Artificial infestation of mites in worker brood:







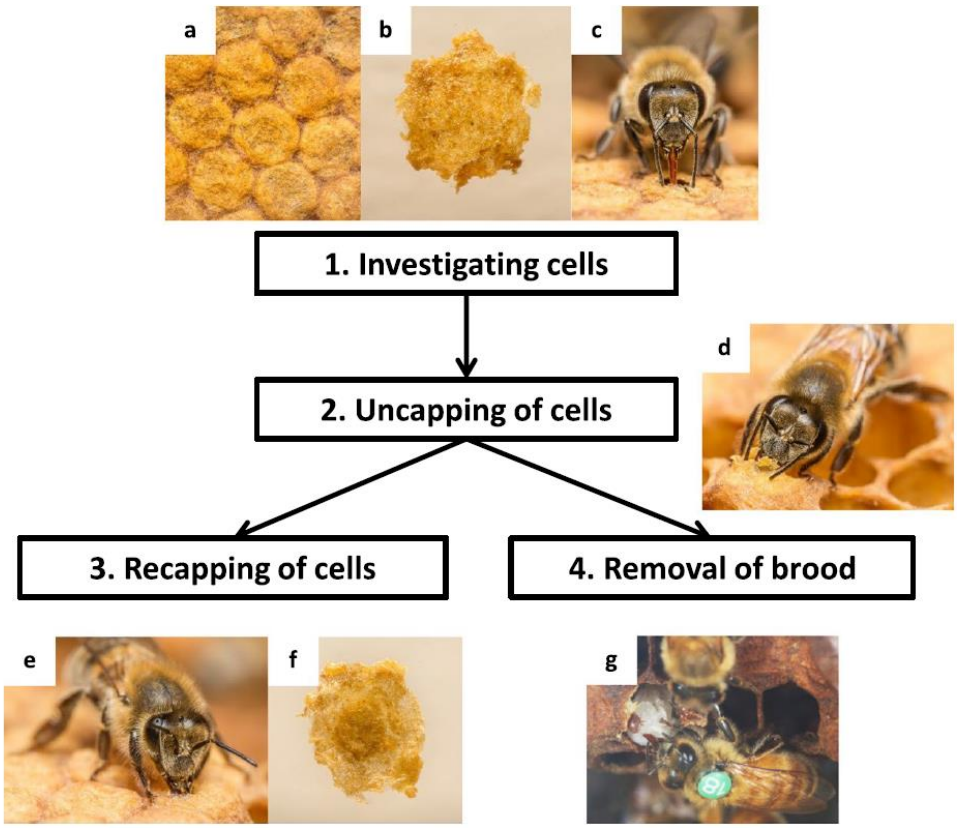


Panziera et., 2017

Population	% removed brood
Control	24
Tiengemeten	15
Lelystad (AWD)	40

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Recapping behaviour (Arrigo Moro 2017, 2018)

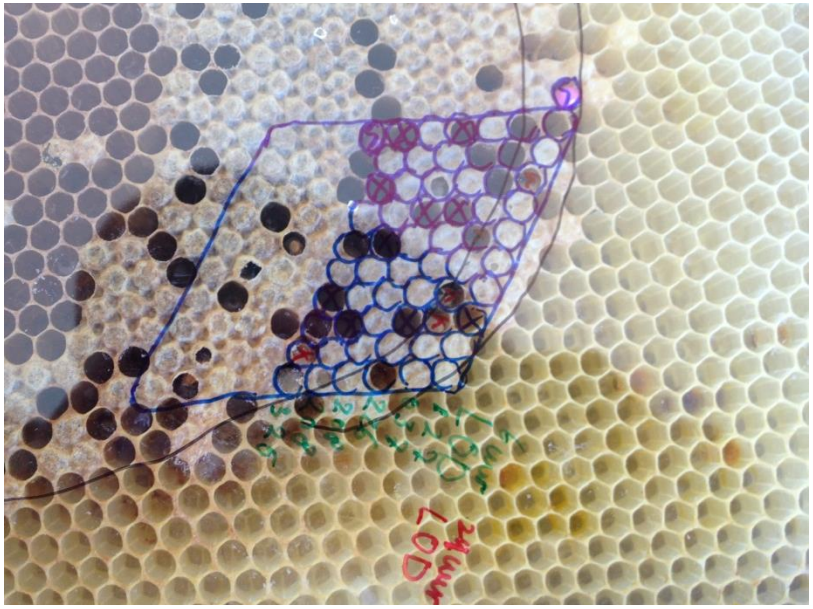


From Oddie et al., 2018

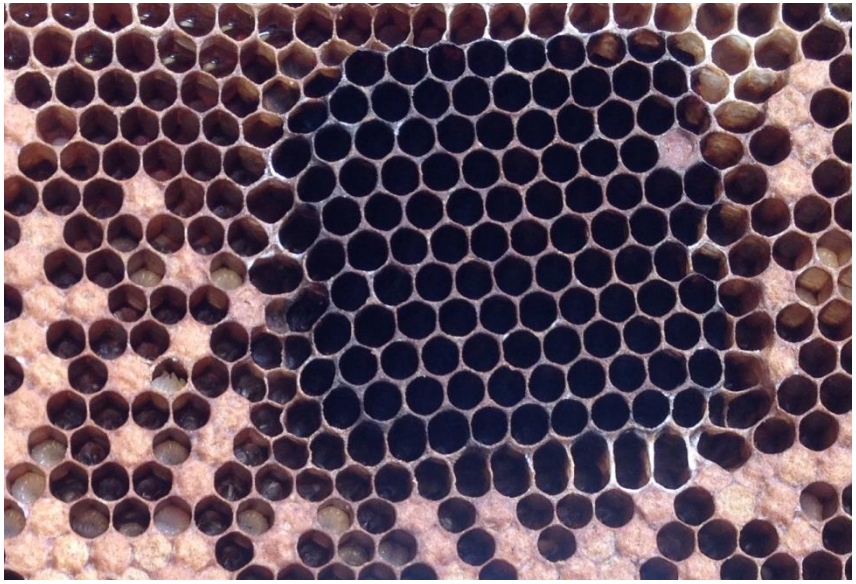
- Recapping behaviour proposed as a less costly trait for the colony
- No difference between groups in 2017 and 2018
- Observed VSH decrease between 2015 and 2017-2018
- Lower fecundity

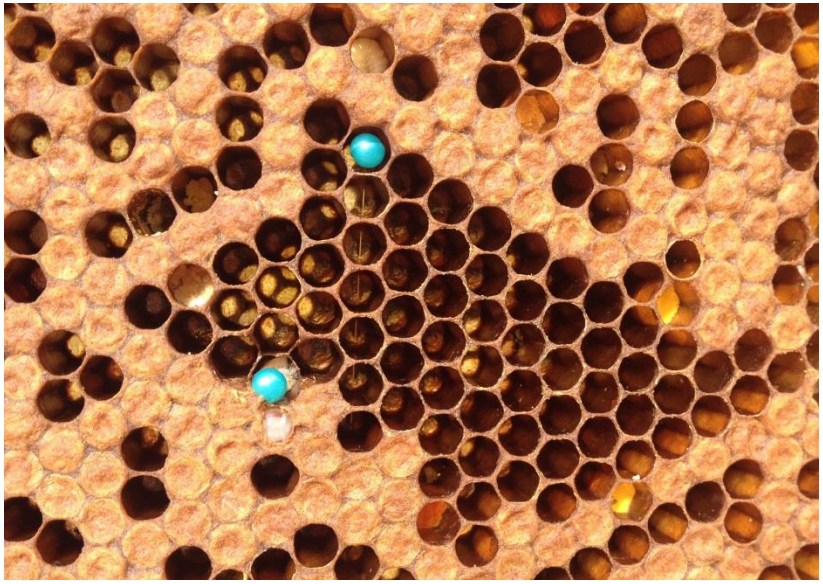
‘Pin test’ and ‘Frozen brood test’





Dead brood (frozen) or wounded brood (pin) as a proxy for Varroa sensitive hygiene





Results of the 'pin test' (Arnout Berenhout, 2016)

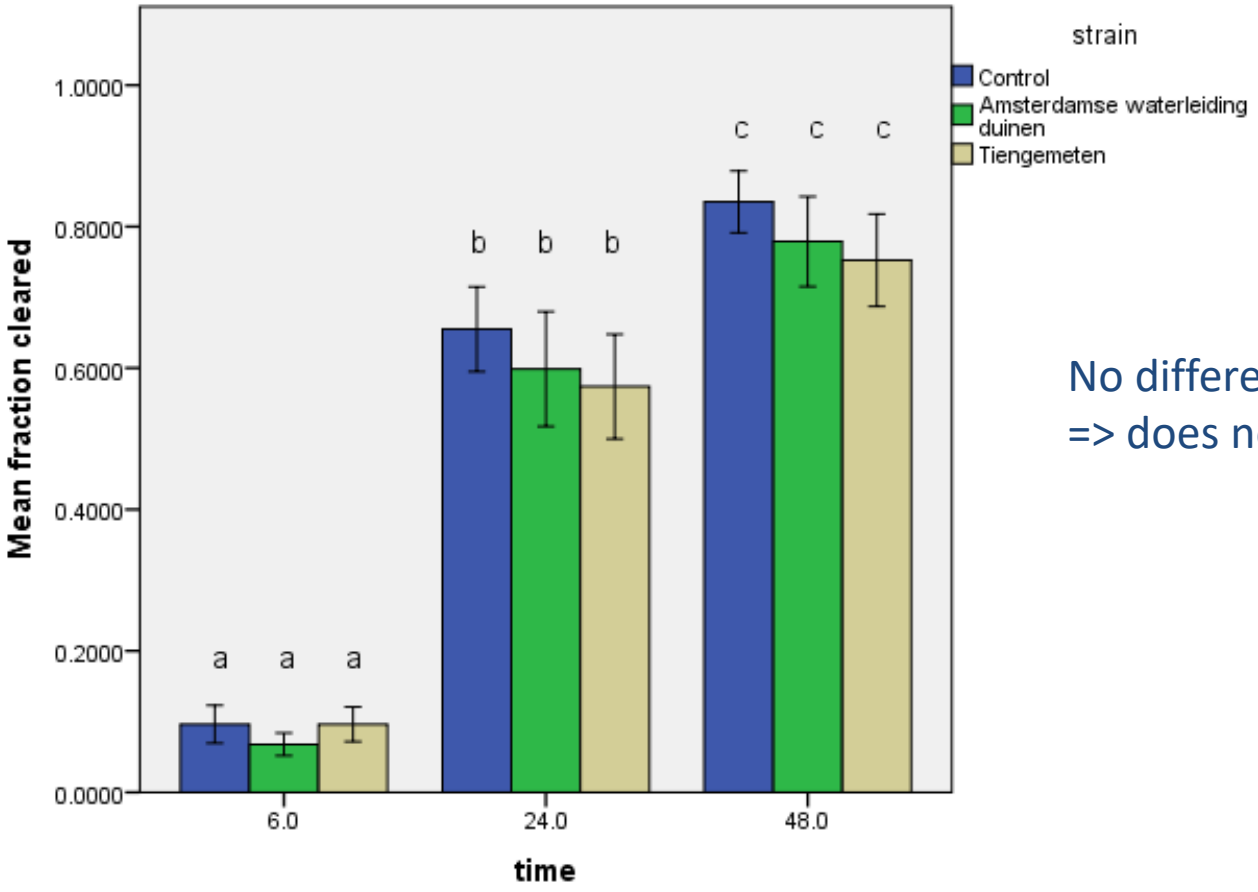
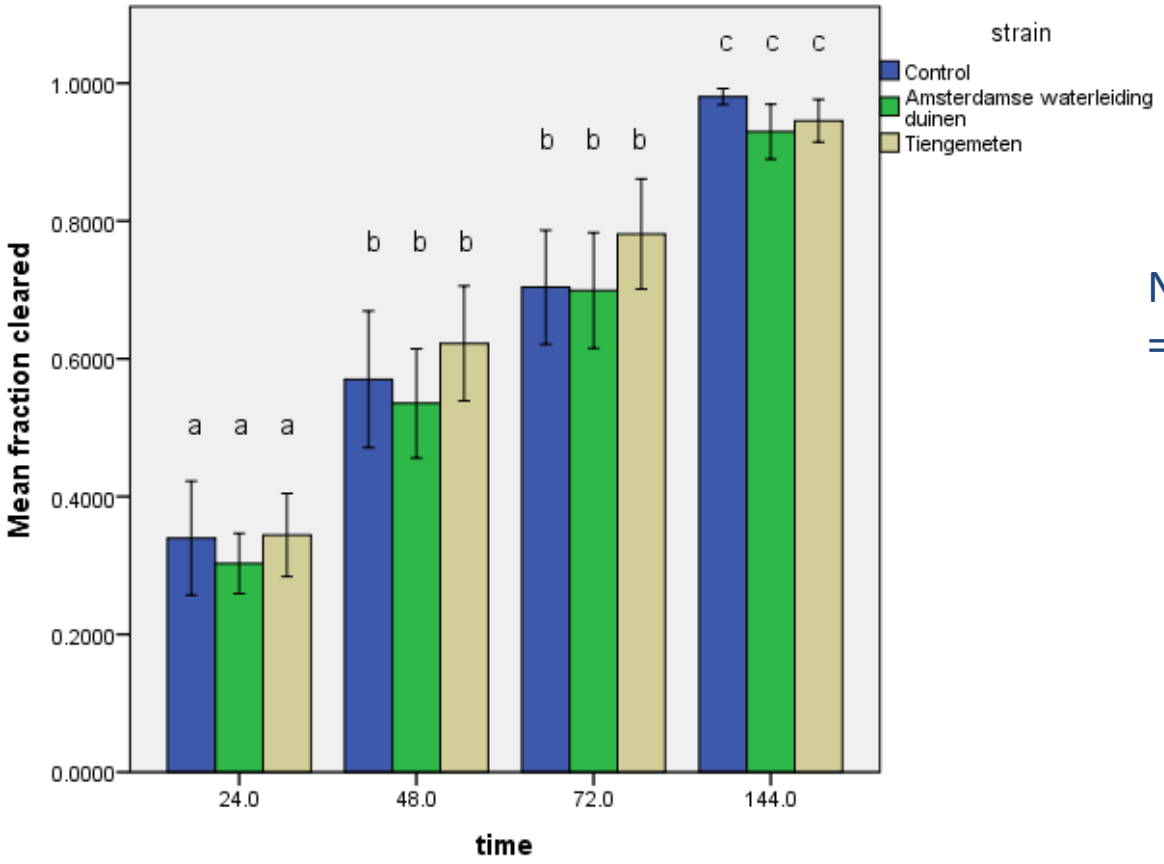


Figure1. Mean (+/- 1 SE) fraction cleared cells, 6, 24 and 48 hours after Pin-test by 6 colonies of each strain.

No difference between groups
=> does not correlate with VSH

Results of the 'frozen brood test' (Arnout Berenhout, 2016)



No difference between groups
=> does not correlate with VSH

Pin and frozen brood tests are **not appropriate** methods for measuring Varroa-specific hygienic behaviour.

Virus tolerance (Barbara Locke 2021)

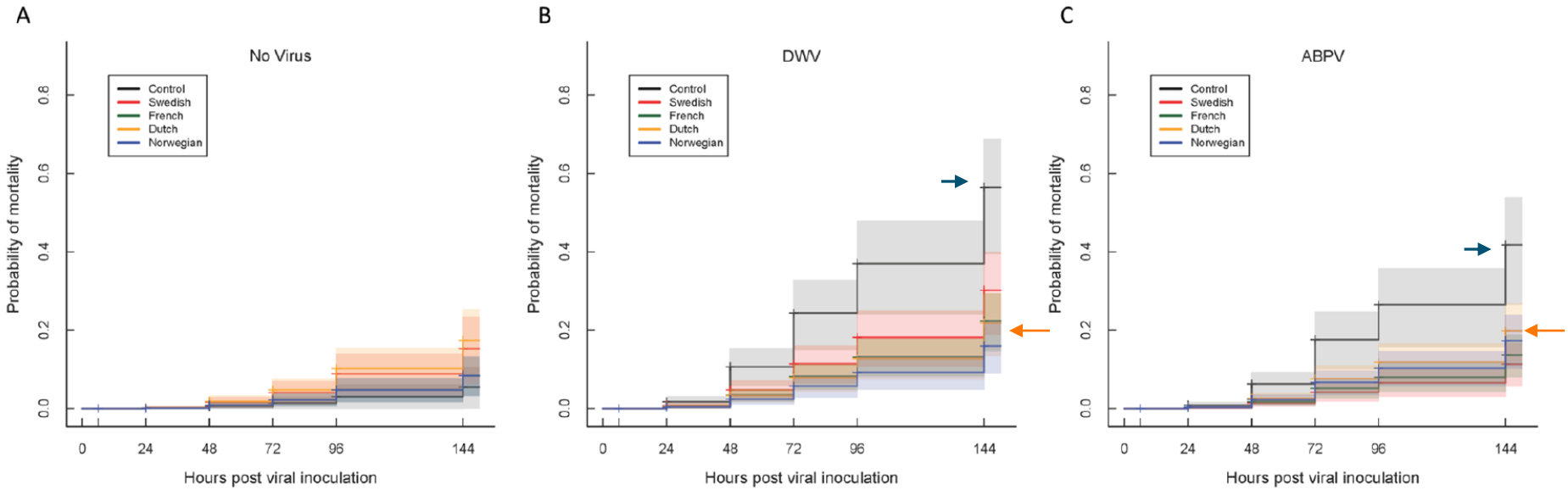
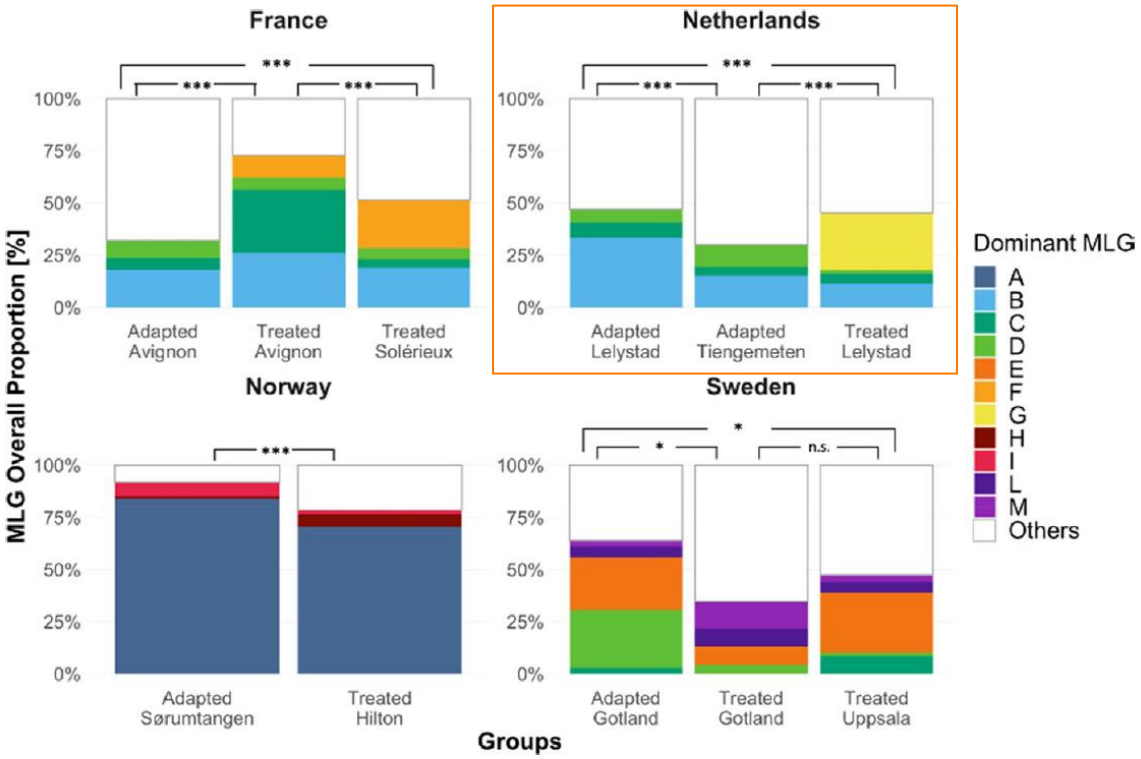


Figure 2. Cox Proportional Hazard curves for the Dutch, French, Norwegian and Swedish varroa-resistant honeybee populations and the varroa-susceptible Control population for the non-inoculated (**A**), DWV-inoculated (**B**) and ABPV-inoculated (**C**) adult bee virus infection experiments. The shaded areas represent the 95% confidence intervals for the proportional hazard lines, based on the data from four replicate honeybee colonies for each population. The effects of varroa infestation and apiary origin of the colonies have been accounted for in the models.

- Resistant populations have higher tolerance to virus

Selection on mites (Arrigo Moro and Alexis Beaurepaire, 2020)



- Selection on mites is strong, since colonies from different populations have different mite genotypes even though they share the same apiary all year round except for the mating period.

Summary

Mecanism	Important for resistance in our populations?
Grooming	No
Varroa sensitive hygiene (VSH)	Yes (L)
Reduction of mite fertility (non- reproduction)	Yes (T)
Reduction of mite fecundity (number of offspring)	Yes (T)
Recapping behaviour	No?
Virus tolerance	Yes
Selection on mites	yes



The importance of these traits might vary over time

New selection populations





Netherlands (2018): Flevo Polder - Zeewolde, 46 colonies

Belgium (2019): Kalmthout, 46 colonies

Germany (2018): Burscheid, 51 colonies

France (2021): Savoie, Auvergne, Normandie



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ARTICLE

Check for updates

A Plea for Use of Honey Bees' Natural Resilience in Beekeeping

Tjeerd Blacquière and Delphine Panziera



- *Apis mellifera* is endemic
- Bees belong as wild pollinators
- Cavity availability might be a limit (NL)



Wild-living colonies are present in beech forests in Germany



From Kohl PL, Rutschmann B. (2018) The neglected bee trees.





The Bed & Breakfast project

In three locations:

- Fairly remote (for the NL) and with ample forage

Boxes design based on Seeley data:

- Red cedar wood of 3.5 cm thickness
- Volume of ~40-45 L
- Narrow entrance
- Mounted in trees ~5 m high

Set up donor apiary nearby:

- Resistant population colonies
- Allow swarming











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The Bed & Breakfast project: locations











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The Bed & Breakfast project







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The Bed & Breakfast project

	2019		2020						2021			
VOLK	22 aug	31 okt	17 mrt	14 mei	17 juni	1 juli	4 aug	30 sep	30 mrt	14 mei	22 juli	11 aug
D-1	+	+	+	+	+	+	-	-	-	+	+	+
D-2	+	+	+	+	+	+	+	+	+	+	-	-
D-3	+	+	+	+	+	+	+	-	-	+	+	+
D-4	+	+	+	+	+	+	+	+	+	+	+	+
D-5	+	+	+	+	+	+	+	+	+	+	-	-
I												
II												
III	+	+	-	-	+	+	+	+	+	+	+	+
IV												
V	+	+	+	+	+	+	+	-	-	-	+	+
VI												
VII												
VIII												
IX	+	-										
X												
XI												
XII						+	+	+	+	+	+	+
XIII	+	+	+	+	+	+	+	+	+	+	+	+
XIV	+	+	+	-	-	+	+	+	+	+	+	+
XV												

- 4 wild-living colonies survived winter
- 1 new swarm has established in the boxes
- 1/3 of boxes occupied
- all were sampled

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Genetic diversity

- *Apis mellifera* genetic diversity has dramatically declined since the 1950's

SCIENTIFIC REPORTS
nature research
Check for updates

OPEN

Declining genetic diversity of European honeybees along the twentieth century

Gonçalo Espregueira Themudo[1,2,3], Alba Rey-Iglesia[1,4,8], Lucía Robles Tascón[5,8], Annette Bruun Jensen[6], Rute R. da Fonseca[5,7] & Paula F. Campos[1,4]

The European honeybee (*Apis mellifera*) is a key pollinator and has in the last decades suffered significant population decline. A combination of factors, including decrease in genetic diversity and introduction of *Varroa* mites, have been suggested to be responsible for these losses, but no definitive cause has yet been appointed. In Europe not only have wild colonies been severely affected, but managed hives have had a massive decline in numbers. To test the hypothesis that honeybees' genetic diversity has decreased in the recent past, we used reduced representation genome sequencing of 40 historical honeybee specimens collected in Natural History collections across Europe and compared them to genomic data from 40 individuals from extant populations (collected post 2006). Our results are consistent with the existence of five evolutionary lineages as previously described, and show a decrease in genetic diversity between historical and extant individuals of the same lineage, as well as high levels of admixture in historical specimens. Our data confirm that a loss of genetic diversity has occurred during the last century, potentially increasing honeybees' vulnerability to contemporary ecological and anthropogenic stressors.

Possible causes:

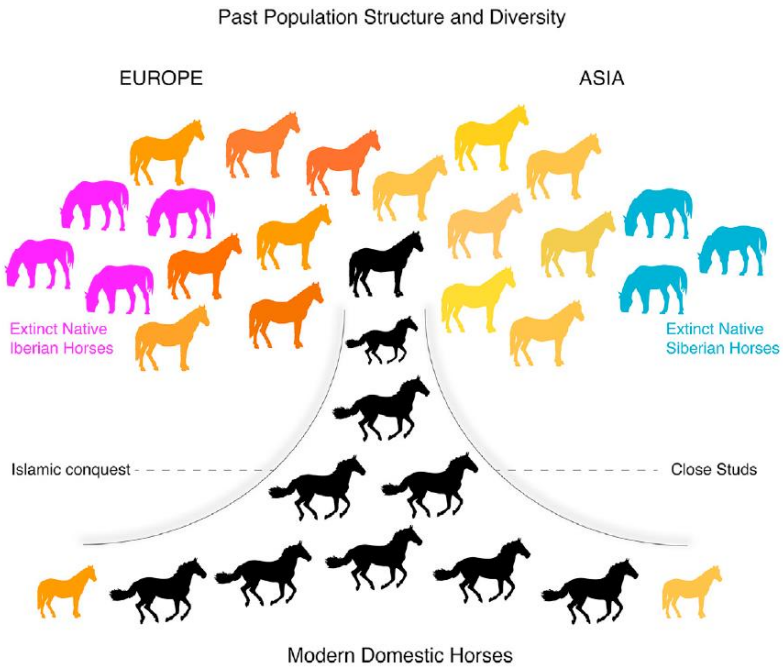
- artificial selection pressure
- lower density of colonies
- habitat fragmentation

Espregueira Themudo, Gonçalo, et al. "Declining genetic diversity of European honeybees along the twentieth century." *Scientific reports* 10.1 (2020): 1-12.



Domestication and diversity

- Honeybees are not domesticated (yet)



Domestication is strongly correlated with:

- Loss of functional traits (horse)=> loss of local adaptations (Kovačić et al., 2020)

Fages, Antoine, et al. "Tracking five millennia of horse management with extensive ancient genome time series." *Cell* 177.6 (2019): 1419-1435.
Kovačić, Marin, et al. "Effects of selection and local adaptation on resilience and economic suitability in Apis mellifera carnica." *Apidologie* 51.6 (2020): 1062-1073.



Domestication and diversity

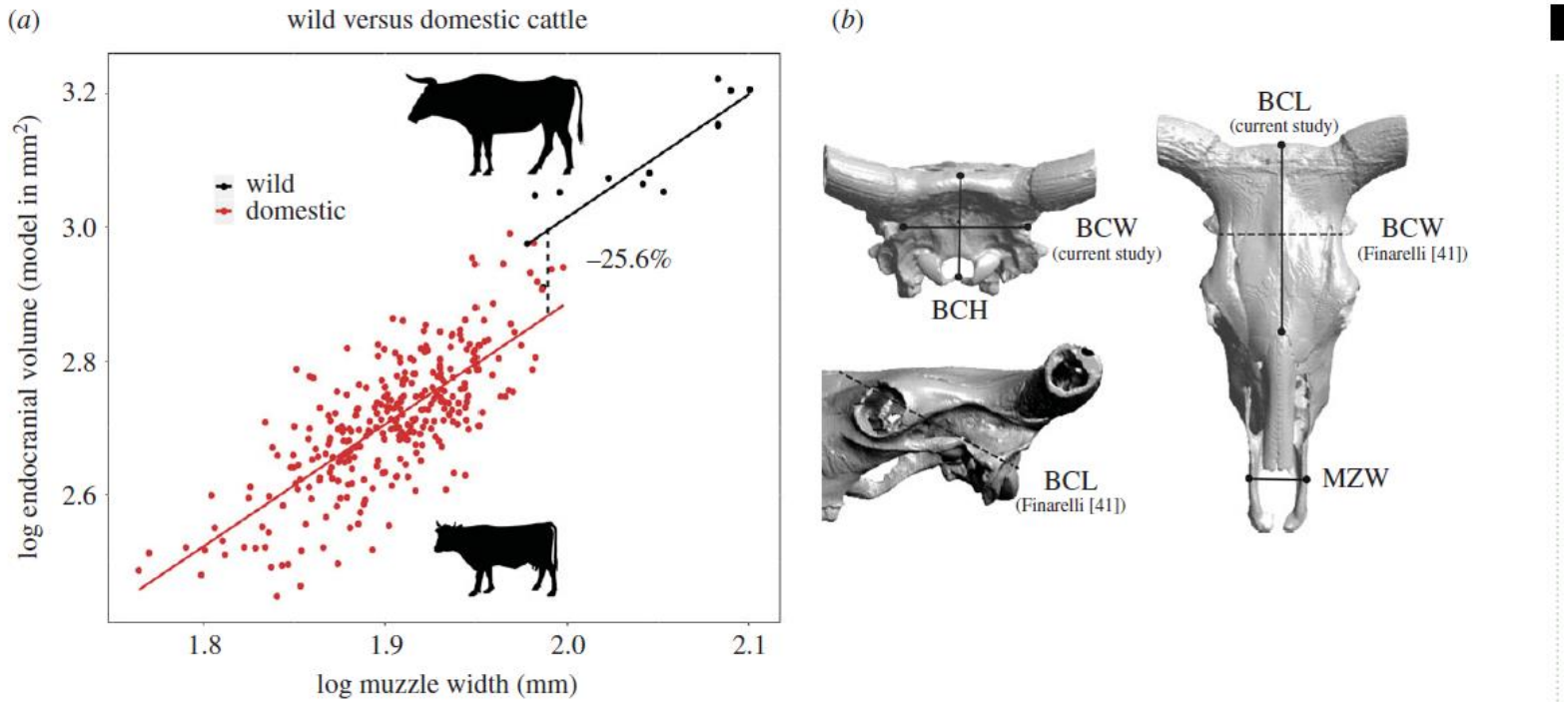


Figure 1. (*a*) Relative brain size difference between wild and domestic cattle. Regression of EV estimate versus MZW; table 1 for statistics. (*b*) Skull measurements

Domestication is strongly correlated with:

- Loss of brain volume (Cattle)=> loss of 'hive wisdom'- flexibility in division of labour, limitation to reach homeostasis (Oldroyd and Fewell, 2007)

Balcarcel, A. M., et al. "Intensive human contact correlates with smaller brains: differential brain size reduction in cattle types." *Proceedings of the Royal Society B* 288.1952 (2021): 20210813.
Oldroyd, Benjamin P., and Jennifer H. Fewell. "Genetic diversity promotes homeostasis in insect colonies." *Trends in ecology & evolution* 22.8 (2007): 408-413.



Domestication and diversity

- Lower diversity (1 patriline per colony vs 15) affects colony growth and survival (Mattila and Seeley, 2007)

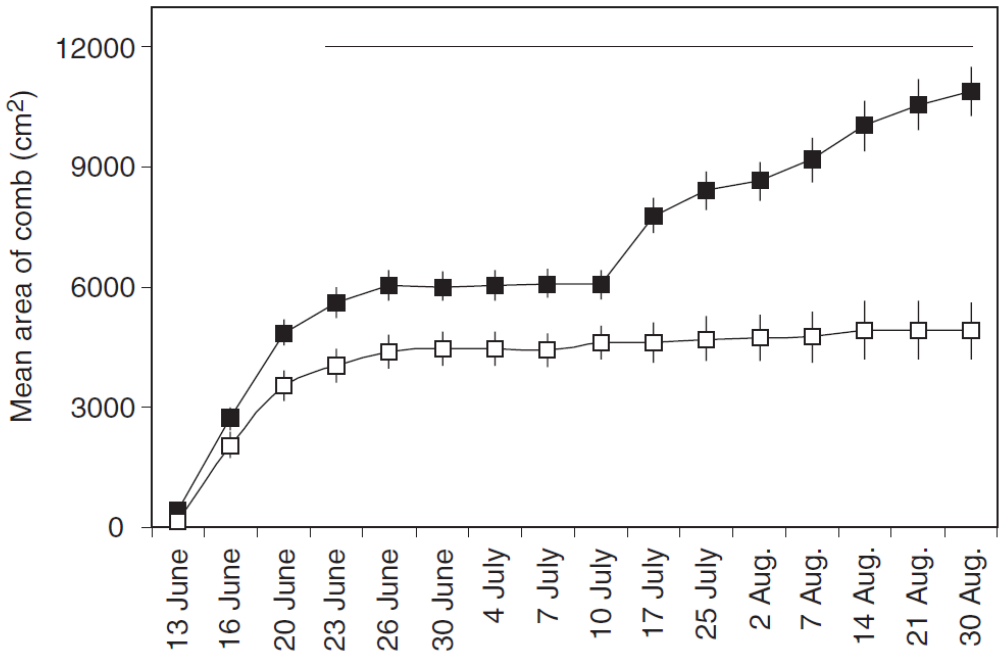


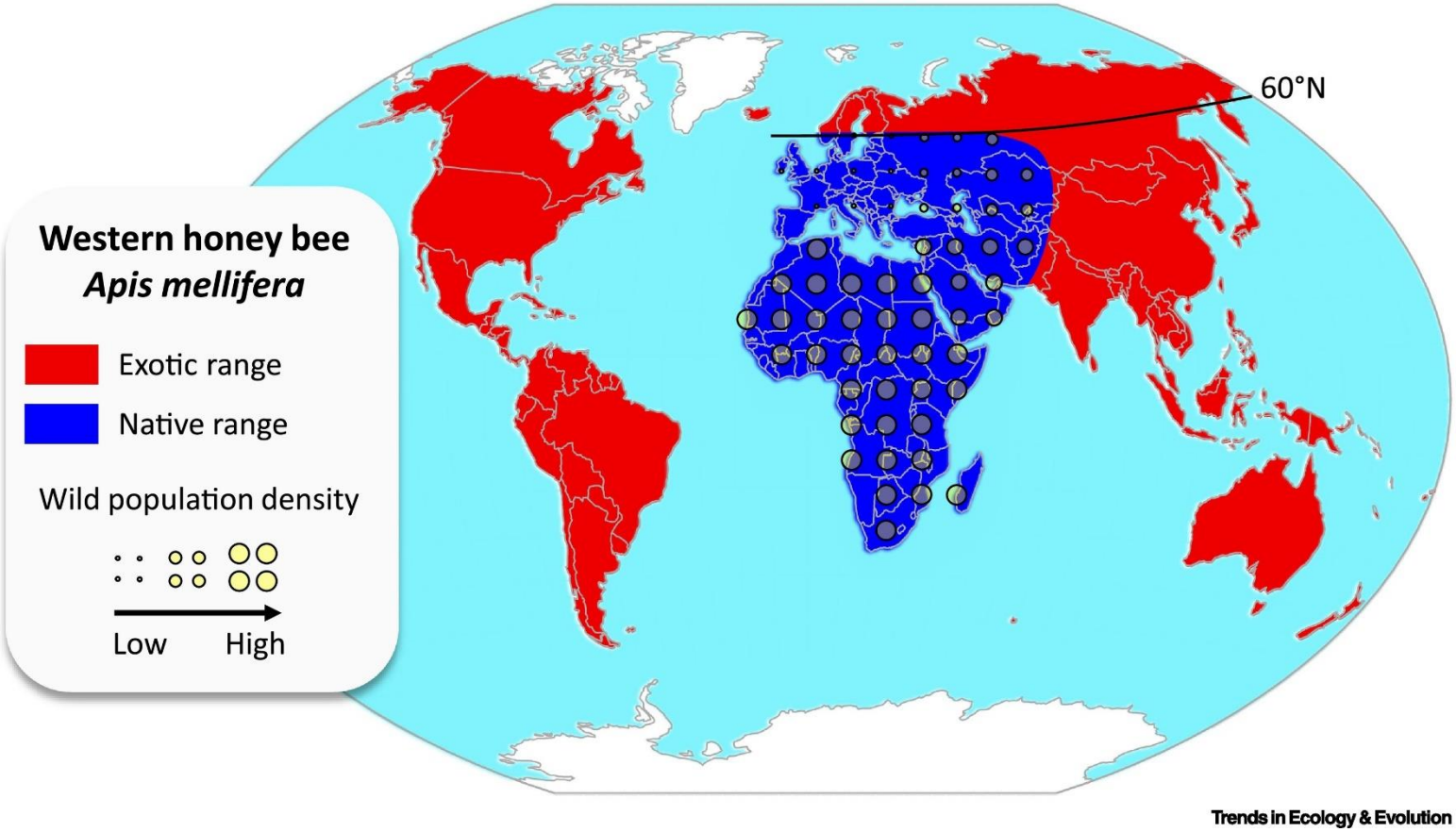
Fig. 1. Area of comb (means ± SEM) constructed by genetically diverse (■) and genetically uniform (□) colonies after occupying new nest sites on 11 June. Dates when groups differed significantly in comb area are indicated by a horizontal line (top).

Mattila, Heather R., and Thomas D. Seeley. "Genetic diversity in honey bee colonies enhances productivity and fitness." *Science* 317.5836 (2007): 362-364.



Merging beekeeping and conservation

- Conserving genetic diversity is crucial
- Increase of effective population size = increase of functional diversity



Requier, Fabrice, et al. "The conservation of native honey bees is crucial." *Trends in ecology & evolution* 34.9 (2019): 789-798.



Merging beekeeping and conservation

- **Conserving the species diversity** is crucial for the long-term sustainability of beekeeping
- **Coexistence of wild-living** and managed colonies in which natural selection is allowed



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Thank you!