Genetic Parameters Of New Traits To Improve The Tolerance Of Honeybees To Varroa Mites

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Introduction

Honeybees (*Apis mellifera*) are primarily known as honey producers. However, the economic value of honey production plays a minor role compared with the economic value of honeybees as pollinators of crops (e.g. Gallai at al. (2009)). But the number of honeybee colonies has dropped during the last few decades in Europe and the US. Winter losses of up to 30% have concomitantly been reported, one reason being the ectoparasitic mite *Varroa destructor*, a major threat to beekeeping. Apiculturists use acaricides to protect their colonies but, now that the first acaricide-resistant mites have been reported, the development of new methods to combat *Varroa destructor* is urgently required. Breeding for Varroa resistance seems to be a promising alternative to medicinal treatment; however, traits characterising resistance against this parasite are difficult to achieve. For several years, honeybee populations in several countries in Europe have been subject to genetic evaluation by using a BLUP model (Bienefeld at al. (2007)).
This contribution aims at estimating genetic parameters for two additional traits used to characterise Varroa resistance in performance testing in the German honeybee population.

Material and methods

Data recording. During the last 15 years, several traits have been recorded within the German *Apis mellifera carnica* population in order to establish reliable selection criteria for Varroa tolerance. Two traits have now been selected to predict Varroa tolerance: the population growth within the performance testing period within the colonies (V1) and the percentage of pierced brood cells removed by workerbees (V2), see www.toleranzzucht.de.

- ***Trait V1:*** *Varroa population growth* is estimated as the ratio of the number of dead Varroa mites found (for 3 weeks) at the bottom of hives in the spring to the number of mites found in a sample of worker bees at the end of the season. This number of mites is adjusted for the sample size of bees (mites per gram). A ratio of both measurements is transformed to fit the normal distribution, see figure 1.
- ***Trait V2:*** The *rate of uncapping pierced brood cells* (pin test) is related to the hygienic behaviour of worker bees; 50 capped brood cells are pierced within a marked area of a brood comb and, approximately 8 hours later, the percentage of removed cells is recorded. The test is repeated three times within the testing period.

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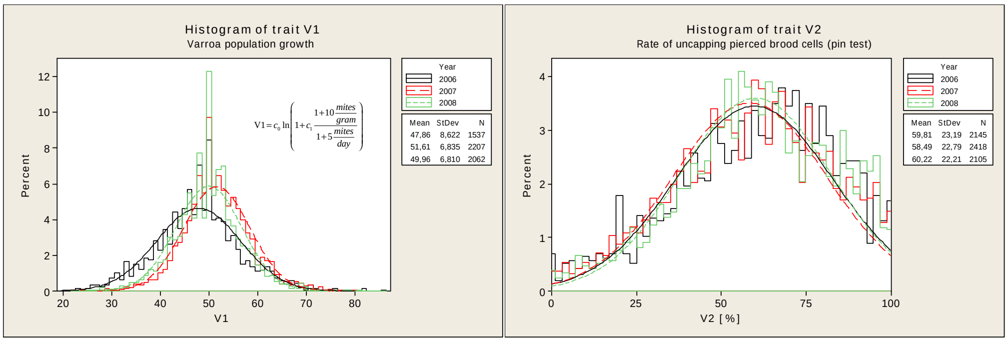
Statistical analyses and transformation. A summary of all collected data of both traits and the transformation of trait V1 is given in figure 1.

Figure 1: Annual distributions of traits V1 and V2



Estimation of genetic parameters. The *AIREMLF90* code provided by Misztal at al. (2002, 2008) has been adapted to the peculiarities of honeybee genetics. Since the controlled pairing of a virgin queen is organised in Germany mainly at isolated mating stations (mainly islands), all successful drones are descended from a group of sister queens. Because of the complex situation at mating stations, only the mother of a queen and the grandmother of the drones that she mates with are listed in the pedigree. To generate a suitable pedigree, we insert a dummy father into the numerator relationship matrix representing the sister group, which provide the drones the queen mates with. According to Bienefeld et al. (1989) the genetic relationship between and within colonies can be estimated by

$$fS(q,n) = \frac{2q+n+qn-1}{q+n+3qn-1}; \qquad hS(q,n) = \frac{2q+3n+qn-2}{4(q+n+3qn-1)}$$

where the relationship of full siblings fS and of half siblings hS is given as a function of the number q of the drone-producing colonies at the mating place and the average number n of haploid drones per queen.

The usual approach for the computation of theoretical breeding values $\mathbf{u_i}$ (e.g. Mrode and Thompson (2005)) for the offspring of parents (*s=sire*, *d=dam*) is here modified to

$$\mathbf{u_i} = \tfrac{1}{2}\mathbf{u_d} + p\mathbf{u_s} + \left[\tfrac{1}{2} - p(q,n)\right]\overline{\mathbf{u}}_\mathbf{s} + \mathbf{m_i}, \qquad p(q,n) = \tfrac{1}{2}\left(2(fS(q,n) + hS(q,n)) - \tfrac{1}{2}\right)^{0.5}$$

with $p < \frac{1}{2}$, whereby $\overline{\mathbf{u}}_\mathbf{s}$ denotes the average breeding value of the father's generation. The paternal (dummy father - offspring) path coefficient $p(q,n)$ is estimated by a least squares approach (Bienefeld et al. (2007)).

Computation of the variance of the Mendelian sampling $\mathbf{m}_\mathbf{i}$ results in an additional term compared with the conventional approach

$$var(\mathbf{m_i}) = var(\mathbf{u_i}) - var(p\mathbf{u_s}) - var(0.5\mathbf{u_d}) - 2\,cov(p\mathbf{u_s}, 0.5\mathbf{u_d})$$
$$= \sigma_u^2\left[1 - p^2(1+F_s) - 0.25(1+F_d) + (1-2p)F_i\right],$$

whereby F_s, F_d and F_i are rates of inbreeding of the dummy father, of the mother, and an average worker bee. Whereas the inverse of the numerator relationship matrix $\mathbf{A}$ is usually decomposed to $\mathbf{A}^{-1} = \mathbf{M}^{-1} \cdot \mathbf{D}^{-1} \cdot (\mathbf{M}')^{-1}$, the elements of the diagonal matrix $\mathbf{D}$ are now defined, as

$$\mathbf{D} = diag\left(var(\mathbf{m_i})/\sigma_u^2\right) = \left\{\begin{matrix} 1 \\ 1 - p^2(1+F_s) \\ 1 - 0.25(1+F_d) \\ 1 - p^2(1+F_s) - 0.25(1+F_d) + (1-2p)F_i \end{matrix}\right\}$$

depending on the number of known parents, whereby σ_u^2 is the additive genetic variance of the trait. The conventional formula results in the case of $p=0.5$ only.
For both traits the model includes a fixed apiary effect, random direct (W=worker) and maternal (Q=queen) effects

$$\mathbf{y} = \mathbf{Xb} + \mathbf{Z_W u_W} + \mathbf{Z_Q u_Q} + \mathbf{e}$$

to consider the combined contribution of the queen and the worker bees to the colony performance.

Results and discussion

Table 1: Genetic parameters for Varroa traits in honeybees[α]

Genetic parameters of Varroa traits		Varroa population growth		Hygienic behaviour	
		W	**Q**	**W**	**Q**
Varroa pop. growth	**W**	**0.32**±*0.03*	*-0.63±0.04*	*-0.25±0.07*	*-0.80±0.03*
	Q		**0.17**±*0.02*	*-0.12±0.07*	*0.89±0.02*
Hygienic behaviour	**W**			**0.47**±*0.02*	*-0.13±0.07*
	Q				**0.15**±*0.03*

[α] Direct and maternal heritabilities (± S.E.) of each trait on the diagonal, and genetic correlations above the diagonal. W=worker (direct effect), Q=queen (maternal effect)

The heritabilities of the worker and of the queen effect in both traits gives hope for the successful breeding for Varroa resistance. As for many other species and traits, negative genetic correlations between direct and maternal effects are found. Negative genetic correlations hinder the genetic response and have to be considered in genetic evaluation. In the case of Varroa tolerance, which is composed of two traits, the genetic correlations between direct and maternal effects between the two traits has also to be considered. Additionally, an estimate of the eventually different “economic” importance of the two Varroa traits is required. These “economical” weights have been estimated by a partial regression analysis of both traits to the survival time of selected colonies with no treatment against the parasite.

The corresponding breeding values are available at www.beebreed.eu. However, most honeybee breeders still focus on traditional traits such as honey yield, gentleness or swarming drive.

Conclusion

A new protocol has been established to characterise the growth of the population of Varroa mites within honeybee colonies. This trait and the recording of the hygienic behaviour of colonies are the basis for the breeding for Varroa tolerance. In each trait the heritabilities are found to be sufficient but the selection response is hindered by negative genetic correlation between direct and maternal effects. A BLUP approach based on both these traits and the genetic correlations between the direct and maternal effects between the traits should provide a promising tool for the successful breeding for tolerance.

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References

Bienefeld, K., Reinhardt, F. Pirchner (1989). *Apidologie* 20:439-450.
Bienefeld, K., Ehrhardt, K., Reinhardt, F. (2007). *Apidologie* 38:77-85.
Gallai, N., Salles, J.-M., Settele, J., Vaissiere, B. E. (2009). *Ecol.Econ*. 68:810-821.
Misztal, I., Tsuruta, S., Strabel, T. *et al.* (2002). In *Proc 7th WCGALP*, vol. 28:7-8.
Misztal, I (2008), *J. Anim. Breed. Gen.*, 125 (6):363-370.
Mrode, R.A., Thompson, R. (2005). Linear models for the prediction of animal breeding values. 2nd ed. CABI-Publ. Wallingford, UK.