

626. Very local genotype by environment interaction in Austrian honey bees

E.W. Brascamp^{1*}, M. Rubinigg^{2,3}, R.F. Veerkamp¹ and P. Bijma¹

¹Wageningen University & Research, P.O. Box 338, 6700 AH Wageningen, the Netherlands; ²Biene Österreich, Georg-Coch Platz 3/11a, 1010 Vienna, Austria; ³CARE, Appalachian State University, ASU, Box 32049, Boone, NC 28608, USA; pim.bascamp@wur.nl

Abstract

We investigated genotype by environment interactions (G×E) in the breeding programme of the Austrian Carnica population administered by Biene Österreich by the estimation of genetic correlations (r_G) at three levels: between geographical regions, between colonies that were bred at home or elsewhere, and between individual test locations. Traits included were honey yield, gentleness, calmness and swarming behaviour. Results suggest that honey bees are very prone to G×E, not only between regions and bred at home v. elsewhere, but also at the level of individual test locations. Despite this abundance of G×E we conclude that breeding programmes covering larger areas are justified, and will result in selection for general adaptability.

Introduction

Between 2009 and 2012 a Europe-wide experiment was carried out to study genotype by environment interaction (G×E) in honey bees (Costa *et al.*, 2012). It was triggered by the phenomenon of colony losses (Neumann and Carreck, 2010), partly associated with Varroa infestation, and the experiment intended to clarify whether G×E played a role in vitality and performance of colonies. To evaluate G×E, queens that were fertilized at their location of origin were distributed across locations in Europa and introduced in host nuclei. An overview of the published results was given by Meixner *et al.* (2014). G×E was found for vitality, performance and behavioural traits, and for vitality colonies performed best at their local environment. This led to the notion of local adaptation, a long-term adaptation to the local environment by selection. Presently the notion of local adaptation is dominant in thinking about honey-bee genetic improvement and conservation of local breeds.

Despite the evidence of G×E there are breeding programmes that cover larger geographical areas, like the whole of Germany and Austria and a few other countries in the case of Beebreed (Hoppe *et al.*, 2020), Austria in the case of Biene Österreich (Brascamp *et al.*, 2016) and Switzerland (Guichard *et al.*, 2020).

Here we investigate G×E in the breeding programme of the Austrian Carnica population administered by Biene Österreich by the estimation of genetic correlations (r_G) at three levels: between geographical regions, between colonies that were bred at home or elsewhere, and between individual test locations. We briefly discuss the consequences of our findings for breeding-programme design.

Materials & methods

The data contained 16,924 records on honey yield, gentleness, calmness and swarming behaviour, collected in the breeding programme of Biene Österreich from performance testing year 1998 to 2020. Honey yield was weighed, and the other traits were scored on a scale of 1-4. Full pedigrees were available. For details, see Brascamp *et al.* (2016) with data until 2014.

Austria recognizes eight 'landwirtschaftliche Hauptproduktionsgebiete' (main agricultural production areas, MPA). The MPA in which colonies were tested, were known for all records. MPA8 differs from

MPA1-7 in an agro-ecological sense, and shows a far higher honey yield. For this reason, we considered two geographical regions, MPA1-7 v. MPA8.

We also distinguished between colonies bred at home v. bred elsewhere. For all records, both the breeder of the queen heading a colony and the tester of the colony were known. When the breeder differed from the tester, the colony was said to be bred elsewhere. In case of colonies tested elsewhere, the identities of queens were blinded to ensure objective measurements, but testers could distinguish own colonies from colonies bred elsewhere. Table 1 summarizes the data.

Methods. We studied G×E by estimating the genetic correlation (r_G) between the traits expressed in one environment v. the other. In honey bees, the consensus statistical model includes two random genetic effects, a worker effect and a queen effect (Bienefeld and Pirchner, 1990). However, after splitting the data into groups for different environments, this model did not converge. We, therefore, included only a worker effect. The numerator relationships matrix was based upon a pedigree file including groups of workers, dams, and groups of drone-producing queens (sires), following Brascamp and Bijma (2014). All models included a fixed effect for year×tester×apiary (called *test location*), and also for bred at home v. elsewhere except when analysis was limited to bred at home. Data were analysed using ASReml (Gogel *et al.* 2015).

To evaluate G×E due to the MPA, we estimated r_G between MPA1-7 and MPA8, either using all records or only the records of colonies bred at home (9,038 for MPA1-7 and 2,703 for MPA8). To evaluate G×E due to bred at home v. elsewhere, we estimated r_G between the trait expressed in colonies tested in MPA1-7, bred at home (9,038) v. elsewhere (4,724).

To evaluate G×E at the level of individual test locations, we defined two hypothetical traits ('between test locations' scenario). We randomly selected half of all test locations present in our data, and allocated all records of these test locations to the first trait. All records of the remaining test locations were allocated to the second trait. When a tester had different test locations within a year, care was taken that these were evenly distributed over the two traits. Consequently, the genetic correlation between the two hypothetical traits originates solely from covariances between test locations, whereas information for genetic variances originates also from covariances between records belonging to the same test location. Hence, an $r_G < 1$ indicates G×E between test locations.

To validate our analysis, we tested whether G×E fully disappeared (i.e. $r_G \approx 1$) when there was no systematic difference between the environments for the two hypothetical traits. For this purpose, we randomly selected half of all records from each test location, and allocated these records to the first trait. The remaining records

Table 1. Numbers of colonies (N), means and standard deviations (between brackets) for colonies tested and bred in different regions (MPA1-7 v. 8).

Tested	Bred	Home/elsewhere	n	Honey	Gentleness	Calmness	Swarming
1-7	1-7	Home	9,038	39.1 (24.9)	3.67 (0.49)	3.65 (0.48)	3.66 (0.72)
1-7	1-7	elsewhere	4,724	34.5 (21.7)	3.54 (0.61)	3.54 (0.59)	3.53 (0.89)
1-7	8	elsewhere	174	39.2 (28.3)	3.62 (0.52)	3.61 (0.52)	3.46 (0.93)
8	8	Home	2,703	85.0 (38.3)	3.64 (0.46)	3.61 (0.46)	3.52 (0.85)
8	8	elsewhere	102	75.1 (28.5)	3.66 (0.50)	3.65 (0.54)	3.35 (0.85)
8	1-7	elsewhere	183	36.4 (20.0)	3.59 (0.49)	3.61 (0.50)	3.39 (0.93)

of each test location were then allocated to the second trait. Hence, each test location was split into two, including an equal division of full sib colonies within a test location ('within test locations' scenario). In this analysis, both the information for the genetic correlation and the information for the genetic variances originate from between and within test-location covariances. Hence, this analysis should yield $r_G \approx 1$, which served as a check for spurious $G \times E$.

Results

Table 2 shows results for r_G . Even though the dataset is of considerable size, standard errors on estimated r_G were large. The estimated r_G for honey yield between MPA1-7 and MPA8 was low to moderate, but not significantly different from one. The large standard errors on these estimates indicate a lack of statistical power, and our results should not be interpreted as evidence of absence of $G \times E$. The estimated r_G for honey yield between colonies bred at home and bred elsewhere was small (0.36 ± 0.14), and significantly different from one, indicating clear $G \times E$. When entire test locations were randomly divided over two hypothetical traits, then the estimated r_G between these two traits was significantly different from 1 for all four traits. Thus there was clear $G \times E$ at the level of individual test locations. In contrast, when records within test locations were randomly divided over to the two traits, all estimates of r_G were virtually unity, as expected. Hence, we did not find indications of spurious $G \times E$ in our analysis.

Discussion

Because $G \times E$ might be caused by the skewed distribution of the four traits, we looked at Box-Cox transformed data (Box and Cox, 1964) in the analysis in the full dataset of MPA1-7 v. MPA8. The transformed variables equalled $(y^\lambda - 1)/\lambda$ where y is the observed value. For honey yield λ was 0.3 and for the other three traits it equalled 2. Results were similar to those in Table 2 and for that reason we didn't transform the data for final analyses.

The standard errors for the estimates of r_G were much lower in case test locations were divided randomly over two traits as compared to the dataset with the actual trait groups (Table 2). This was probably caused by the tighter genetic relationships between colonies assigned to both traits as compared to the actual groups.

Results suggested that honey bees are very prone to $G \times E$. Even when dividing test locations arbitrary over two traits, there was considerable $G \times E$ between these traits. This result contrasts with results generally found in farm animals, where r_G is often high.

Genetic correlations between different regions found here were in line with Hoppe (2021, personal communication) who observed a broad range of genetic correlations between regions in the dataset of Beebreed (*re* Hoppe, 2020, for description of the data) using statistical models similar to ours. In our

Table 2. Genetic correlations with SE (between brackets) between regions, between bred home vs elsewhere tested in MPA1-7, between test locations, and within test locations.

Data used	Honey	Gentleness	Calmness	Swarming
MPA1-7 v. MPA8	0.55 (0.32)	-0.31 (0.64)	0.64 (0.28)	-0.51 (0.58) ¹
MPA1-7 v. MPA8, home bred	0.28 (0.61)	0.88 (0.39) ¹	0.84 (0.56) ¹	-0.95 (0.69)
Home v. elsewhere in MPA1-7	0.36 (0.14)	0.68 (0.17)	0.89 (0.28)	0.92 (0.70) ¹
Between test locations	0.63 (0.07)	0.85 (0.07)	0.75 (0.08)	0.41 (0.16)
Within test locations	0.99 (0.01)	0.99 (0.02)	0.99 (0.02)	0.96 (0.06)

¹ This analysis did not result in stable estimates for the additive genetic (co)variance components.

analysis this G×E wasn't statistically significant in terms of r_G , but given the significant G×E when dividing test locations arbitrary over two traits, the null-hypothesis of $r_G=1$ is not realistic, and therefore we consider the G×E between regions as real. This type of G×E may arise because different sets of alleles or allele frequencies may be optimal for different regions and may be expected to remain so over longer periods. On the other hand, for the contrast bred at home *v.* elsewhere one might think of short term effects, perhaps of an epigenetic nature. Yagound *et al.* (2020) demonstrated in honey bees that methylation patterns in paternal germ cells, through drones, were present in worker offspring. In our case it would involve the maternal pathway, as workers in colonies bred at home and elsewhere often share the same sire. The queens heading the colonies of course differed. If G×E for tested at home *v.* elsewhere is a short-term effect indeed, it may diminish over generations. This might have implications for experiments in which different genetic stocks are being tested at home and elsewhere, as in the Europe-wide experiment. Based on our findings, one should consider to evaluate stocks for successive generations, such that in later generations all are tested at home.

For farm-animal scenarios Mulder and Bijma (2006) addressed the question which level of r_G would justify to set up different breeding programmes for different countries or regions, or at least to work with different sets of estimated breeding values. Considering the high degree of G×E in honey bees it seems tempting to set up local or region-specific breeding programmes, suggesting splitting a breeding programme into very many small ones. However, the resulting small scale for each programme would obviously hamper genetic progress and increase rates of inbreeding. Furthermore the estimates of heritability of Brascamp *et al.* (2018), who used largely overlapping Biene-Österreich data and ignored G×E, suggest that genetic improvement across environments is realistic for the population as a whole. Hoppe *et al.* (2020) demonstrated considerable genetic improvement in Beebreed as well, where breeding value estimation ignores G×E.

We conclude that G×E is abundant in Austrian honey bees, but that this provides no basis to split breeding programmes into smaller ones, and that the solution lies in selection for general adaptability.

References

- Bienefeld K., and Pirchner F. (1990) *Apidologie* 21:175–183. <https://doi.org/10.1051/apido:19900302>
- Box G.E.P., and Cox D. R. (1964) *J. R. Stat. Soc. B* 26:211–252.
- Brascamp E.W., and Bijma P. (2014) *Genet. Sel. Evol.* 46:53. <https://doi.10.1186/s12711-014-0053-9>
- Brascamp E.W., Willam A., Boigenzahn C., *et al.* (2016) *Apidologie* 47(6):739–748. <https://doi.org/10.1007/s13592-016-0427-9> and (2018) *Apidologie* 49(4):462–463. <https://doi.10.1007/s13592-018-0573-3>
- Costa C., Büchler R., Berg S., *et al.* (2012) *J. Apicultural. Sci.* 56(1):147–158. <https://doi.org/10.2478/v10289-012-0015-9>
- Gogel B.J., Gilmour A.R., Welham S.J., *et al.* (2015) *ASReml Update: What's new in Release 4.1*. VSN International Ltd, Hemel Hempstead.
- Guichard M., Neuditschko M., Soland G., *et al.* (2020) *Apidologie* 51(5):876–891. <https://doi.org/10.1007/s13592-020-00768-z>
- Hoppe A., Du M., Bernstein R., *et al.* (2020) *Insects* 11:768. <https://doi.org/10.3390/insects11110768>
- Meixner M.D., Büchler R., Costa C., *et al.* (2014) *J. Apicultural Res.* 53(2):183–187. <https://doi.10.3896/IBRA.1.53.2.01>
- Mulder H.A., and Bijma P. (2006) *J. Dairy Sci.* 89:1727–1739. [https://doi.org/10.3168/jds.S0022-0302\(06\)72241-X](https://doi.org/10.3168/jds.S0022-0302(06)72241-X)
- Neumann P., and Carreck N. (2010) *J. Apicultural Res.* 49:1–6. <https://doi.org/10.3896/IBRA.1.49.1.01>
- Yagound B., Remnant E.J., Buchmann G., and Oldroyd B.P. (2020) *PNAS*, 117(51):32519–32527. <https://doi.10.1073/pnas.2017094117>