Modelling the impacts of imports of non-native honey bees into the native *Apis mellifera mellifera* population in Ireland

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1 Abstract

The human-mediated import of non-native queens is threatening the unique adaptations of native honey bees in Ireland through hybridisation and introgression. To study the impacts of this import, we built a SIMplyBee stochastic model of hybridisation between a native *Apis mellifera mellifera* population in Ireland and a non-native (C lineage) honey bee population. The model included colony honey yield and fitness as complex polygenic traits. We modelled the effects of varying the percentage of imports, the genetic correlations between fitness in native and non-native environment, and the spatial distribution of introgression over 20 years. We measured the levels and rate of introgression and the changes in genetic means for honey yield and fitness. The results revealed that increased imports expectedly increase the rate of introgression. This introgression induced a trade-off between increased honey yield and decreased fitness. Changing the genetic correlation between fitness in native and non-native environment pointed to a larger fitness decrease with negative correlations. The study showed the spread of introgression across the whole simulated area, indicating that even colonies far from the import zones could be affected over time. Furthermore, halting imports generated a desirable direction of change, however the purging of introgressed material was very slow and varied substantially across replicates. These results underscore the risks and long-term impacts of import on complex polygenic traits. We argue that the best way forward to manage the problem of import and associated introgression is to invest in holistic selective breeding of the native honey bee population in its native environment rather than import invasive subspecies that are bred to be successful in a different climate. The study also highlights the lack of fundamental quantitative genetic parameters for complex polygenic traits of different honey bee populations in different environments.

2 Introduction

The Western honey bee *Apis mellifera* is the most widely recognised bee species worldwide due to its extensive distribution and commercial use [31]. The species is highly stratified into 7 mitochondrial lineages and 33 subspecies identified, each characterised by factors such as morphometry, behaviour, molecular approaches, and differential trait expression [11, 31]. A desired trait among beekeepers is honey yield due to its high commercial value. Two subspecies of the C lineage (*A. m. ligustica* from Italy and *A. m. carnica* from Slovenia) are popular for commercial beekeeping globally [30, 44]. These subspecies have been the focus of intense breeding to generate colonies that show high honey yield, gentle nature, low propolis usage, and high fertility [55]. For these reasons, beekeepers around the world import queens of these subspecies. However, such imports are contributing to the decline of locally adapted populations through the introgression of their non-adaptive genetic material and the possibility of introducing new diseases [32, 18].

Introgression is the transmission and integration of genetic material from one species, subspecies, or lineage into the genetic background of another with continued hybridisation and backcrossing [18]. Introgression can be associated with an increase in genetic diversity, which can boost the evolutionary potential in changing environments [32]. However, it can also lead to the loss of unique combinations of alleles that have emerged over long periods of adaptation, and can hence change adaptive traits in a population [59, 6]. Persistent imports and introgression also pose a risk of genomic extinction of a lineage or a subspecies, which in turn could result in the general homogenisation of the species [42, 14].

A notable example of the effects of import and introgression in recent times, is the reduction of the natural range of *A. m. mellifera*. This subspecies is found in northern Europe and belongs to the M lineage and it is raising concerns among researchers, beekeepers, and beekeeping associations in places right across Europe including the Nordic countries,

UK, and Ireland [28, 17, 52]. Beekeepers from Ireland are reporting problems with the maintenance of their native population of *A. m. mellifera* due to increased imports of the queens of C lineage in recent years [41]. In the UK, 20.000 queens were imported in 2020 [34]. As a consequence, native honey bee populations of Ireland and the UK exhibit varying degrees of introgression [32, 18, 27, 45, 41]. This introgression can change the unique traits of *A. m. mellifera*, such as the ability to mate in poor weather conditions [41]. Some suggest that if imports continue, *A. m. mellifera* could disappear from both of these countries [32].

Some *A. m. mellifera* populations in Norway, Denmark, Sweden, Scotland, and Ireland are still intact, due to their isolation in remote locations making them important reservoirs for this subspecies [24, 48]. In fact, Norway and Denmark have included *A. m. mellifera* in conservation programmes and have banned the import of non-native subspecies into some areas [60]. In Ireland, the Native Irish Honey Bee Society (NIHBS), the Irish Bee Foundation (IBF), the IBA and the Federation of Irish Beekeeping Associations (FIBA), advocate for the protection of their native population and signed an all party statement of the need to ban honey bee imports into Ireland [23].

To gain more information about the current status of *A. m. mellifera*, introgression is actively studied in Ireland and the UK [18, 27, 41]. Unfortunately, there is scarce information about how introgression affects the genetic variation, fitness, and honey production of native populations. Simulations are a valuable tool for a time- and cost-effective evaluation of long-term effects of introgression, and hence a prediction of populations' evolution under such conditions. The results of such simulations could provide insights into how a control or ban of the imports should be implemented in practice. Such simulations are widely used in animal and plant genetics research [54, 19, 2], and are also gaining traction in honey bee research [5, 53, 35, 8, 51, 65].

The aim of this contribution was to build a stochastic simulation model for quantifying the magnitude and effects of introgression in native honey bee populations due to imports of

non-native queens. Using the model, we simulate the Irish native *A. m. mellifera* population and study introgression in different scenarios and quantify the impact of the introgression on honey production and fitness.

3 Material and Methods

The stochastic simulator SIMplyBee [an R package; 51] was used to simulate a natural yearly cycle of honey bee colonies with import of queens from a non-native population. The R code for this simulation is open-source and available in the supplement and on-line (Supplementary material: 8). We simulated two populations that resembled a native Irish population of *A. m. mellifera* and a non-native Central European population of C lineage. Two complex polygenic traits representing fitness and honey yield were simulated in these populations, but with a genetic correlation between the expression of these two traits in the two environments of the populations, giving rise to genotype-by-environment (GxE) interaction. We simulated 10 years of a burn-in phase followed by 10 years of scenarios varying the percentage of imported queens, the magnitude of GxE interaction for the fitness trait, and the point of entry of the imports. We compared the scenarios based on the level of introgression and the genetic mean for fitness and honey yield.

3.1 Simulation

We simulated the founder genomes with the Markovian Coalescent Simulation implemented in SIMplyBee, according to *A. mellifera* demographic model [70, 51]. We generated 500 *A. m. mellifera* and 500 *A. m. carnica* diploid genomes for the founders of each of the native populations. We generated 15 replicates of these base populations. Each genome contained 16 chromosomes with 1000 segregation sites per chromosome. From the founder genomes, we created the base populations of 500 *A. m. mellifera* colonies that represented the population

in Ireland, and 500 *A. m. carnica* colonies that represented the population of in Central Europe providing the source of imports of C lineage queens. Each colony was simulated with one queen and 10 workers, which, to reduce computing time, represented the full size of a colony.

Within each native environment, we simulated two complex polygenic traits, fitness and honey yield. We simulated both traits as effected by 100 additive quantitative trait loci (QTLs) per chromosome (1600 QTLs per trait in total) and a trait mean of 0. For honey yield, we set genetic variance to 0.25, and residual variance to 0.75, which resulted in a heritability of 0.25, as estimated for natural honey bee populations [40]. For fitness, we set the genetic variance to 0.1 and residual variance to 0.9, resulting in a heritability of 0.1, typical of fitness-related traits [38, 22]. We modelled the GxE interaction by simulating fitness and honey yield in both environments as genetically correlated traits, resulting in four simulated traits: honey yield in Ireland, honey yield in Central Europe, fitness in Ireland, and fitness in Central Europe. Therefore, each simulated individual had a genetic and phenotypic value for each of the four traits, regardless of their physical location. However, only the "local" values were used for selection, breeding, or import decisions. For honey yield, we fixed the genetic correlation between environments to 0.75, following Hatjina et al. [25] that showed that honey bees produced more honey in their native environment than in a non-native one. For fitness, we varied the genetic correlation between the scenarios as described in the following.

3.2 Burn-in

During the burn-in phase, we kept the two populations closed and simulated 10 years of a beekeeping yearly cycle within each population. This cycle simulated three beekeeping seasons: spring, summer, and winter, with colony events occurring at different probabilities depending on the season (Table 1). These probabilities were estimated from the current

literature [39, 62, 58, 9, 21]. We ran the 15 replicates of the base populations through the burn-in phase.

In the spring period (Figure 1 A), we first simulated the growth of existing colonies to their full size. Next, we split all colonies to simulate swarming prevention, which generated new colonies that were re-queened with age 0 virgin queens obtained from the original colonies. Then, we swarmed or superseded a percentage of colonies (Table 1). The remnant colonies of the swarms or supersedures were naturally re-queened with their own virgin queens. Next, we generated drones from colonies with queens of age 1 or older to create a drone congregation area (DCA) and mated all the new virgin colonies with an average of 15 random drones from the DCA. Finally, we ended the spring period with the collapse of a proportion of colonies, mimicking queen loss during a nuptial flight (Table 1).

In the summer period (Figure 1 B), we again swarmed a proportion of colonies (Table 1), though less than in the spring period, since swarms occur mostly in spring, when the colonies outgrow their hive [67]. We followed this by superseding or collapsing a percentage of colonies (Table 1). The resulting virgin colonies mated as in the spring period.

Finally, in the winter period (Figure 1 C), we collapsed a larger percentage of colonies due to overwintering challenges (Table 1). We collapsed colonies with the lowest phenotypic values for fitness in the corresponding environment to simulate *directional* selection, driving genetic differentiation between the two populations evoling in two different environments. As a result, the Irish population had a higher genetic mean for "fitness in Ireland" than the Central European population at the end of the burn-in phase. At the end of each yearly cycle, we removed some splits to maintain constant population sizes. In the Central European population, we removed the colonies with the lowest phenotypic value for honey yield to simulate directional selection on this trait. This selection reflected the long-term selective breeding for this trait and the presumed superior honey yields of the imported bees into Ireland.

Table 1: Probabilities of colony swarming, supersedure, and collapse in spring, summer, and winter simulated periods

	Spring	Summer	Winter
Supersede	0.05	0.05	-
Swarm	0.05	0.01	-
Collapse	0.10	0.10	0.18

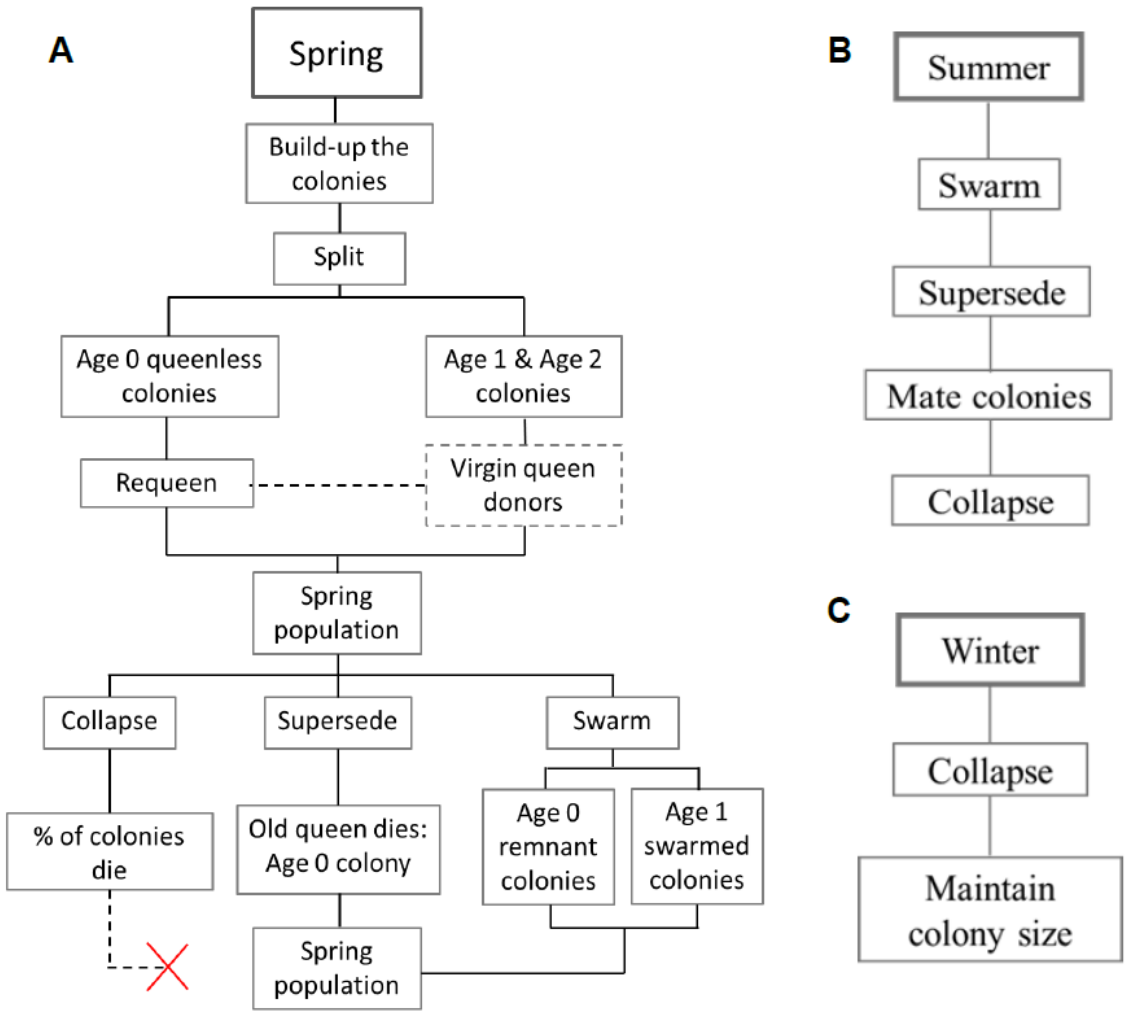

Figure 1: Flow diagram of the simulated yearly cycle of honey bee colonies in A) the spring period, B) the summer period, and C) the winter period

3.3 Scenarios

After the burn-in, we continued running the 15 replicates for each of the three sets of scenarios for 10 additional years. In this phase, we transitioned from directional to stabilising selection on fitness, assuming that both populations had reached their optimal fitness levels after the burn-in. To implement this, we selectively collapsed colonies whose phenotypic value for fitness deviated most from the population's mean fitness during the winter period.

First, we simulated the import of queens from the C lineage into the Irish population by requeening a percentage of spring splits with imported virgin queens (Figure 2). We compared the scenarios with 0% (Control), 1.5%, 4%, and 10% of imports, which were determined based on the reported imports to Ireland [41].

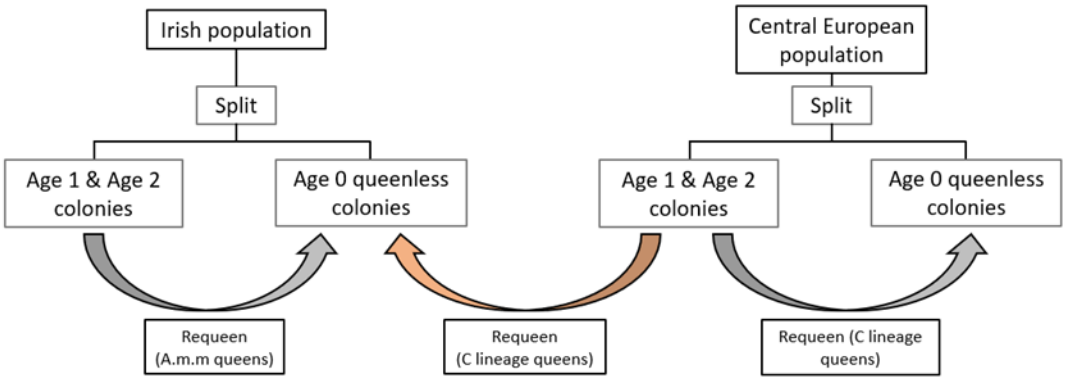

Figure 2: Flow diagram of the import process: re-queening of age 0 queen-less colonies in the Irish population with C lineage queens from the Central European population after the burn-in phase

Second, to explore the effects of adaptation to the local environment and GxE interactions, we created a set of scenarios that varied the genetic correlation between fitness in Ireland and Central Europe. Büchler et al. [10] found higher survival of honey bees with native genotypes over non-native genotypes, indicating the presence of GxE interactions and a past adaptation to the environment. However, given the lack of studies on the extent of the GxE interaction in honey bees, we evaluated a wide range of genetic correlations: -0.75,

-0.25, 0.25, and 0.75 (base scenario).

Third, to explore the effect of the spatial distribution of imports, we created spatially-aware scenarios. In the scenarios described above, the colonies have no notion of the location and can hence mate randomly (panmictic simulation). This was done to simplify the simulations, as we were mostly interested in the qualitative differences between scenarios. However, colony locations and the locations of imports are likely to impact the introgression. SIMplyBee enables spatially-aware simulations, where each colony is assigned a location and the mating is restricted to a user-defined radius. To evaluate the effect of the point of import on the spread of introgression over time and place, we created a scenario by simulating an area with a single point of import at the center of the simulated area. We chose to place the point of import at the center of the simulated area to measure the effects of introgression symmetrically in all directions. We simulated an area of $150km^2$ with 500 colonies randomly placed. This mimicked the colony density in the Dublin county [15], which receives the most of imports in Ireland (about 3,000 reported honey bee colonies within $900km^2$ in 2020).

The 500 colonies were randomly placed into groups of between 5 and 15 colonies, representing the Irish apiaries. We restricted the mating to $15km$ radius and the new location of the swarms to $1.5km$ radius [33, 61]. Colonies created as splits were given the same location as the colony they originated from. Virgin colonies were re-queened with virgin queens sampled from a nearby area. For this, we divided the area into 16 smaller areas and sampled one colony as the virgin queen donor for each area.

Lastly, we simulatedd the effect of halting the import to evaluate if a population can recover after long periods of continuous imports. For this, we extended the spatially-aware scenario with a single point of entry for an additional 10 years (30 simulated years in total), where we halted imports to evaluate if the population can recover after long periods of continuous imports.

3.4 Analysis

To assess the level of introgression, we tracked recombination of genomes throughout the simulation and with it identity by descent inheritance of alleles since the two base populations. This enabled quantifying the proportion of alleles that originate from the native *A. m. mellifera* or the non-native C lineage population. We measured the level of introgression for each queen as 1 minus the proportion of alleles that originate from the founder *A. m. mellifera* alleles. Then we averaged the level over all queens by the year of birth.

We computed the rate of introgression as the slope of a linear model fitting the mean level of introgression over years for each replicate: $lm(\text{Introgression} \sim \text{Year} - 1)$. We omitted the intercept, because the level of introgression was 0 at the end of burn-in. We computed the average slope across the replicates of the scenarios, as well as the 95% quantiles.

We computed the genetic trend for "honey yield in Ireland" and "fitness in Ireland" traits by calculating the mean genetic value of queens by their year of birth in the Irish and the Central European populations for each replicate. These values were centered to have a mean of 0 in the last year of burn-in, year 10. Then we computed the average across the replicates of the scenarios as well as the 95% quantiles

To assess the contribution of imported queens to genetic gain, we next partitioned the overall genetic trends for honey yield and fitness traits in the Irish population to the contribution of native *A. m. mellifera* and non-native C lineage population with AlphaPart R package [50].

In the spatially-aware scenario, we assessed the spread of introgression by plotting the proportion of introgression against the Euclidean distance of the colonies from the point of import. The Euclidean distance d between a colony located at (x_i, y_i) and the point of entry at $(75, 75)$ (centre of the $150km^2$ area) was calculated as $\sqrt{(x_i - 75)^2 + (y_i - 75)^2}$.

Moreover, we analysed the cline of introgression, to study gradual change across geographical distance. We fitted clines for each year of importation using the Hybrid Zone

Analysis R package [16]. For each year of import, we collected the estimated c value, representing the centre of the cline. We averaged the c values across replicates to obtain the mean c value per year, as well as the 95% quantiles.

We performed ANOVA to assess the significance of differences in the collected quantities. We performed the post-hoc Tukey HSD tests to evaluate the differences between the levels of a specific factor. All of these analyses were performed using the base R packages [56].

4 Results

The results show (1) the effects of varying percentage of imports and the genetic correlation between fitness in two environments on the level and rate of introgression and mean honey yield in the Irish population, (2) the dispersion of introgression with a single point of entry, and (3) effect of halting importation practices. Increasing import resulted in a proportional increase in introgression and honey yield; however, it also resulted in a notable decrease in fitness. Changing the genetic correlation between fitness in two environments did not significantly affect introgression or honey yield, but negative correlations led to a stronger decrease in fitness. Introgression from a single point of entry spread fast through the simulated area over the years. Halting imports led to a decay of the level of introgression of the non-native alleles, but the rate of this decay was low.

4.1 Percentage of imports

The results showed that the level and rate of introgression increased from year 10 to year 20 in all scenarios that imported non-native queens (Figure 3). The increase became significantly more evident with higher import percentages (Supplementary material: Table 3 and 4). We expectedly observed the highest rate of introgression in the scenario with 10% imports (0.013). Here, the level of introgression reached 0.39 by year 20, meaning that 39% of the

population's genome was non-native. Decreasing the percentage of imports decreased the level of introgression, with the population being 18.3% and 7.4% introgressed in year 20 respectively with 4% and 1.5% imports (Table 2). The control scenario had no import and hence retained the level and rate of introgression at 0.

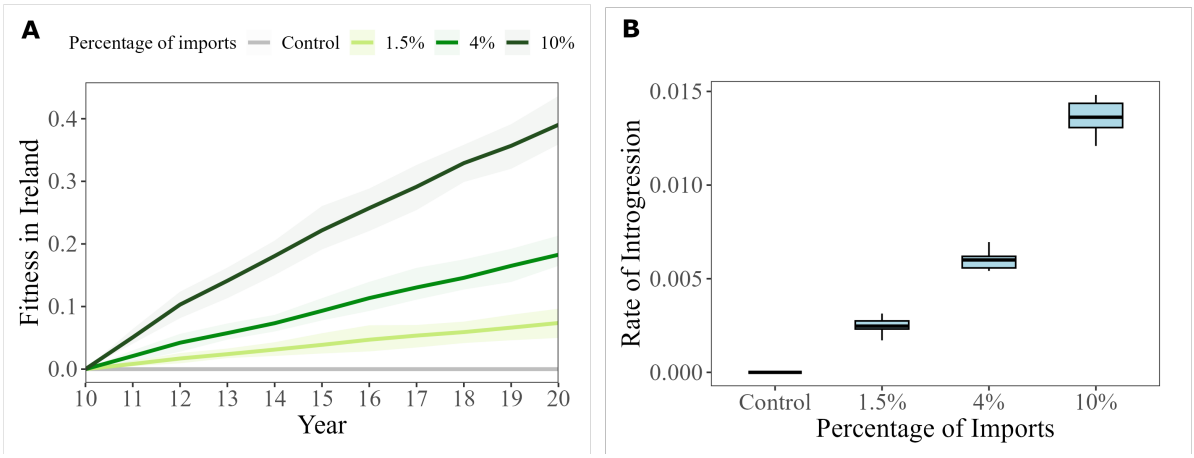

Figure 3: Changes in the level and rate of introgression across replicates in scenarios with the varying percentage of imports of the C lineage into the Irish population: Control (0%), 1.5%, 4%, and 10%. A) The trend in mean level of introgression with the corresponding shaded regions indicating the 95% quantiles. B) Boxplot of the rates of introgression.

Table 2: Values of introgression rates, introgression levels, average honey yield genetic value, and average fitness genetic value in year 20 in the Irish population with varying percentage of imports: Control (0%), 1.5%, 4%, and 10%; and varying correlation between fitness traits in the two environments: +0.75, +0.25, -0.25, -0.75.

	Scenarios	Rate of introgression	Introgression at year 20	Honey yield at year 20	Fitness at year 20
Percentage of imports	Control (0%)	0.000	0.000	-0.008	-0.011
	1.5%	0.002	0.074	+0.041	-0.016
	4%	0.006	0.183	+0.119	-0.041
	10%	0.013	0.393	+0.304	-0.087
Changing genetic correlations	+0.75	0.0061	0.183	+0.119	-0.041
	+0.25	0.0058	0.175	+0.102	-0.066
	-0.25	0.0059	0.176	+0.134	-0.071
	-0.75	0.0057	0.171	+0.138	-0.078

In accordance with the introgression, the mean genetic value for honey yield in the Irish

population increased with increasing percentage of imports, while the mean genetic value for fitness decreased. In the control scenario, traits fluctuated around zero as honey yield in Ireland was not under directional selection, fitness was under stabilising selection at the optima, and there were no imports (Figure 4). We observed the highest increase in mean genetic value for honey yield in the scenario with 10% import, reaching +0.304 genetic standard deviations by year 20. This value was about 7 times higher than in the 1.5% import scenario and about 3 times higher than in the 4% scenario (Table 2, Supplementary material: Table 3). However, the 10% import scenario also showed a significant and steep decrease in mean genetic value for fitness, dropping to -0.087 genetic standard deviations by year 20, while the decrease in fitness in the 4% and 1.5% scenarios were less pronounced and not significantly different from the control group (Supplementary material: Table 4).

The partition of the genetic trends for both traits further confirmed the positive contribution of imported queens to honey yield, with the imports accounting for 99% of the increase in honey yield. This partition also confirmed the negative contribution of imported queens to fitness, with the imports accounting for 99% of the decrease in fitness (Supplementary material: Figure 9 and 10).

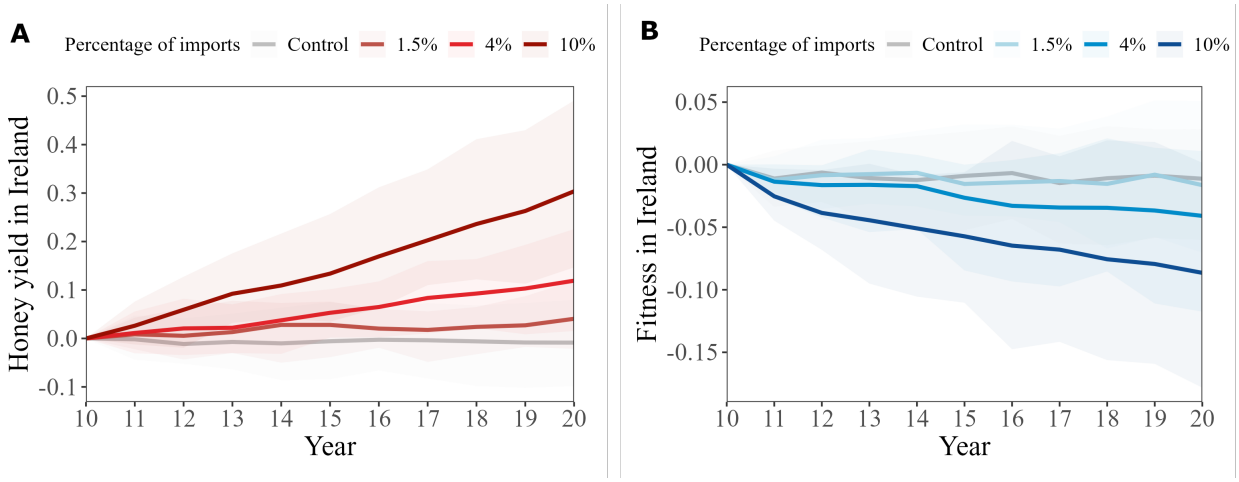

Figure 4: Changes in the mean genetic value for (A) honey yield (red lines) and (B) fitness trait (blue lines) in the Irish population from year 10 to year 20 in scenarios with a varying percentages of imports: Control (0%), 1.5%, 4%, and 10%. The solid lines represent the mean genetic value across replicates with corresponding shaded regions indicating the 95% quantiles.

4.2 Genetic correlation between fitness in the two environments

The results revealed that the genetic correlation between fitness in Ireland and Central Europe had a limited effect on the level and rate of introgression (Figure 5). Although the differences in introgression matched our expectations, there was a substantial variation between simulation replicates and the differences between scenarios were not statistically significant (Supplementary material: Table 3 and 4). The high genetic correlation of 0.75 gave the highest rate of 0.0061/year and the level of 0.183 in year 20. The low genetic correlation of -0.75 gave the lowest rate of 0.0057/year and the level of 0.171 in year 20.

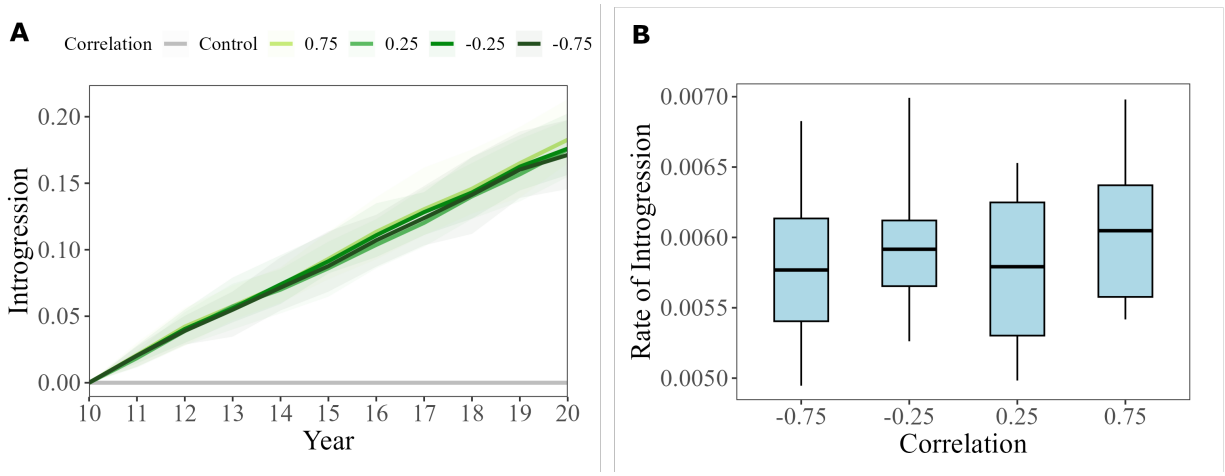

Figure 5: Changes in the level and rate of introgression from year 10 to year 20 across replicates in scenarios with a varying genetic correlations between fitness traits in Ireland and Central Europe: +0.75, +0.25, -0.25, -0.75. The percentage of imports was set to 4% in all scenarios. A control population that received no imports is also represented. A) Trend in the introgression level with corresponding shaded regions indicating the 95% quantiles. B) Boxplot of the rate of introgression.

The mean genetic value for honey yield in the Irish population by year 20 did not follow a pattern regarding genetic correlation for fitness. With the -0.75 correlation scenario exhibiting the highest mean (0.138 genetic standard deviations) and the 0.25 scenario exhibiting the lowest (0.102 genetic standard deviations) (Figure 6, Table 2). However, none of the differences between the scenarios were significant due to the high variation between replicates (Supplementary material: Table 3 and 4).

The partitioning of the genetic trends for honey yield further confirmed that although the import increased the honey yield in all scenarios, there was no clear pattern in the size of contribution related to the genetic correlation for fitness in the two environments (Supplementary material: Figure 11).

The mean genetic value for the fitness trait in the Irish population by year 20 decreased with decreasing the genetic correlation between fitness trait in Ireland and Central Europe (Figure 6, Supplementary material: Table 3). The scenario with the +0.75 exhibited the smallest decrease in the value of fitness by year 20 (-0.041 genetic standard deviations), while

the scenario with the -0.75 correlation exhibited the most significant decrease in the value of fitness by year 20 (-0.078 genetic standard deviations) (Table 2, Supplementary material: Table 4).

The partition of the genetic trend for fitness further revealed the negative contribution of the imported honey bees to the genetic trend for fitness in the Irish population. While the domestic contribution to fitness fluctuated around zero due to stabilising selection, the contribution of the imports was increasingly negative as we decreased the fitness genetic correlation, with the maximum decrease observed with the -0.75 genetic correlation. Fitness declines were mostly driven by the imports, which accounted for 99% of the observed decline (Supplementary material: Figure 12)

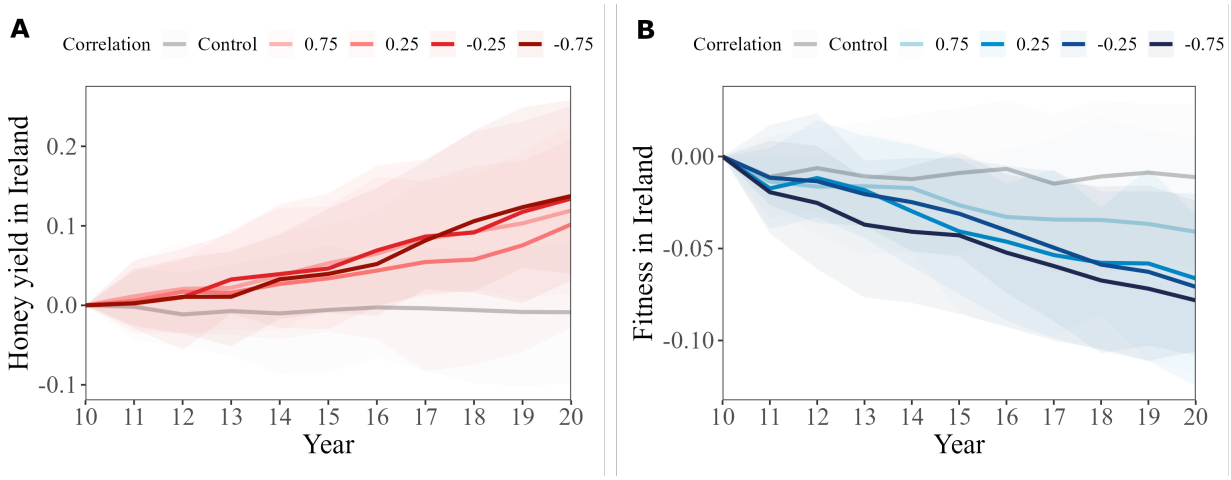

Figure 6: Changes in the mean genetic value for (A) honey yield (red lines) and (B) fitness trait (blue lines) in the Irish population from year 10 to year 20 in scenarios across varying correlations between fitness traits: +0.75, +0.25, -0.25, -0.75. The solid lines represent the mean genetic value across replicates with corresponding shaded regions indicating the 95% quantiles. A control population that received no imports is also represented.

4.3 Point of entry

In the scenario with a single point of entry, colonies closer to the entry point expectedly exhibited higher levels of introgression compared to those further away. For instance, colonies

located in a radius of 25km or closer to the point of entry showed levels of introgression that ranged between 75% to 100%, while colonies 100km away from the entry point showed no introgression over the simulated period (Figure 7a). The extensive spread of C lineage alleles was evidenced by the presence of some level of introgression even in colonies located 80km away from the point of entry at year 20. Moreover, the cline analysis revealed a significant increase in the c value over the years ($p < 0.001$) (Supplementary material: Table 5). Hence, the centre of the cline was moving away from the point of entry, also indicating that introgression was spreading progressively further from the point of entry as years passed (Figure 7b, Supplementary material: Figure 13).

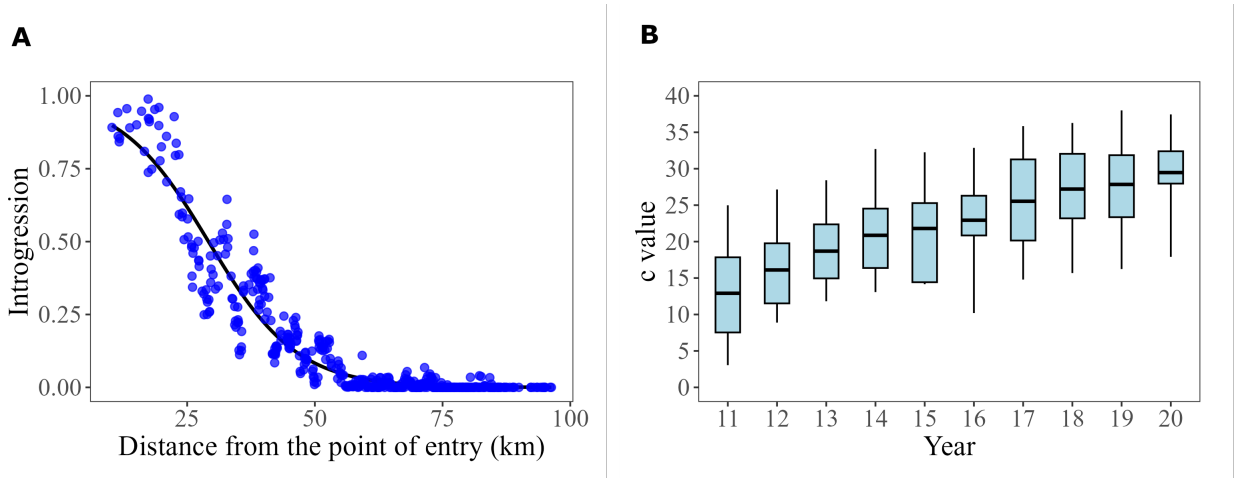

Figure 7: A) Scatter plot of the introgression levels in honey bee queens in relation to the distance from the central point of entry after 10 years of importation. Each blue dot represents a honey bee queen. The black line indicates the fitted cline to the scatter plot. B) Box plot of the annual changes in the c value (centre of the cline), across 10 years of import over replicates.

4.4 Halt of imports

To evaluate the ability of the population to recover from the imports, we extended the spatially-aware scenario (point of entry) by halting imports in year 20 and we continued the simulation for another 10 years.

By year 20, there was a clear increase in the level of introgression, as shown in the results above for the spatially-aware scenario, reaching a value of 0.14 and a rate of introgression of 0.006/year. In the following 10 years without import, we observed a slow decay for the rate of introgression (-0.001/year) compared to the increase observed in the importation period. This led to a gradual decline in the level of introgression, decreasing to 0.13 by the end of the simulation (Figure 8). There was however a substantial variation between replicates on how this level of introgression changed over time.

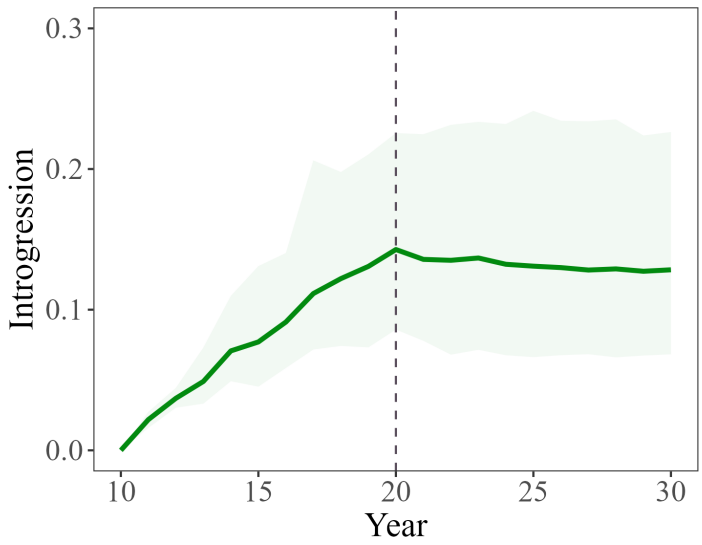

Figure 8: Trend in the mean level of introgression from year 10 to year 30 across replicates with corresponding shaded regions indicating the 95% quantiles. The dashed line marks the halt of import.

5 Discussion

We developed a stochastic model of hybridisation between a native and non-native honey bee populations via import of queens. The model is flexible in terms of major parameters and extendable with additional scripting, hence allowing for the exploration of consequences of imports. The model can be used to study the level and rate of introgression and change

of trait means as a function of published knowledge (such as the percentage of colonies that collapse in each season or the amount of imports) and as a function of yet unknown or debated variables (such as genetic correlations between trait expressions in different environments), which can be treated as a model parameter that varies across scenarios. The model simulates genomes of the two populations from a published honey bee demographic model [70]. It then further generates directional genetic differences between the populations by selecting on traits in different environments during the burn-in phase, which changes the QTL allele frequencies in each population. This way of simulating populations has been used in other breeding and genetics studies [54, 19, 2, 53, 35, 8, 51, 65]. As an extension of these types of simulations, we have also included spatial location of the apiaries and their hives, which enabled us to measure the spread of introgression across the studied area over time. These spatially-aware simulations are enabling a joint study of population genetics and ecology [13].

5.1 Percentage of imports

In Ireland, there have been reports of an increasing number of imported queens in recent years, reaching about 4% in 2012 and higher during covid years [41]. The result of these activities has led to a rise in the level of introgression detected in Irish colonies, reaching 18-25% [69, 63]. Our simulation with this percentage of imports generated 18.3% of introgression after 10 years and the rate of 0.006/year, which is close to the observed values in Irish colonies. While these imports increased the mean genetic value for honey yield, they also decreased the mean genetic value for fitness. Increasing the percentage of imports from 4% to 10% in the simulation expectedly increased the level of introgression, increased the mean genetic value for honey yield, and decreased the mean genetic value for fitness. These results are expected since in the simulation the non-native population was under positive directional selection for honey yield, while the native was not. Also, the non-native population was selected for fitness in their own environment. Under such conditions, the expected contribution of imports to

honey yield and fitness in Irish conditions depends on genetic correlations for these traits between the two environments. Due to imperfect genetic correlation for fitness, imports decreased fitness in the Irish population, however the assumed natural selective pressure with stabilising selection in Ireland was not strong enough to prevent or purge introgression with continued imports. These results are highly dependent on the assumed quantitative genetic parameters and the simulated genetic differences between the populations, as we discuss in the following.

5.2 Genetic correlation between traits in the two environments

Results of the simulation depend critically on the assumed genetic correlation between traits in the two environments. For honey yield we assumed the genetic correlation of 0.75 following Hatjina et al. [25]. For fitness we varied the genetic correlation to study it's impact on introgression. Changing this genetic correlation for fitness substantially affected the mean genetic value for fitness of the Irish population. Changing this genetic correlation from +0.75 to either -0.25 or -0.75 significantly decreased the mean genetic value for fitness in the Irish population. This is expected since a negative genetic correlation between two environments implies that the animals performing best in one environment perform less well in the other environment [e.g. 3]. Therefore, with negative correlations, as the honey bees in Central Europe were increasingly adapted to their environment during the burn-in phase, they were indirectly becoming less adapted for the Irish environment. Although GxE has not been extensively studied in honey bees, there are reports that honey bees show higher survival in their native environment compared to non-native environment [47, 43]. There are also reports that importing non-adapted honey bees could decrease fitness and increase colony losses [10, 29]. However, the amount of GxE in honey bees has not yet been studied in detail, partly due to the lack of sufficiently powered datasets. Such datasets would include a large number of phenotyped and genotyped individuals in different environments and sufficient

genetic connectedness between the environments.

While decreasing the genetic correlation for fitness decreased the mean genetic value for fitness due to imports, we did not observe the expected corresponding reduction in introgression. An explanation for this observations is that the simulated natural selection pressure was not strong enough to purge all non-native alleles. We assumed the Irish population to be at it's fitness optimum and under stabilising selection, but also that fitness is a highly complex polygenic trait affected by 1600 QTLs and has the heritability of 0.1 (hence most of phenotypic variation in fitness is due to environmental effects). Taken together, these assumptions meant that introgressed non-native alleles were effectively largely hidden to selection and could "silently" increase in their frequency over time without major impacts on fitness. Consequently, we did not observe significant differences in the change of mean genetic value for honey yield between scenarios that varied the genetic correlation for fitness. Should we have implemented directional selection for fitness in the Irish population, we expect that purging of non-native alleles would have been more effective, with corresponding impact on both honey yield and fitness.

5.3 Point of entry

The results showed that the impact of honey bee imports into specific sites in Ireland has much greater implications, impacting native subspecies significantly farther from the points of import. After 10 years, introgression was observed in colonies located up to 80km away from the initial point of entry. This distance was influenced by the parameters set in our model, particularly the queen mating radius, which was set to 15km based on Jensen et al. [33]. This meant that queens in each colony could only mate with drones from colonies within 15km, therefore defining the maximum spatial extent of gene flow in a single mating season. This aligns with the natural mating behavior of honey bees, which disperse and mate over large areas in the absence of physical barriers [1, 12].

The majority of honey bee imports in Ireland arrive at ports or major cities, such as Dublin, Belfast, and Cork [69]. Although most of these cities are coastal, we placed the import point in the center of the study area, yet we simulated the area based on the number of colonies found in Dublin [15]. The choice of simulating a central point of entry was made to (1) quantify the spread of introgression without being limited by coastline boundaries, (2) allow for a more generalizable scenario regarding the point of entry, and (3) ensure that sufficient data points were available around the point of entry for accurate analysis. Moreover, the choice of point of entry did not affect the introgression, which was of our main interest. We also assumed that there are no major geographical barriers, such as mountain ranges, within this study area. This largely matches with the geography of Ireland.

Overall, our results suggest that imported non-native alleles can disperse extensively, reaching colonies tens of kilometers away within a decade. This has further implications if imported honey bees are traded within Ireland and distributed to other locations where they further disperse. Future work could refine these predictions made here by incorporating real-world geographic features, varying colony densities, and more complex models of bee flight and mating behavior. For instance, a future upgrade of the model could include differences in the mating behaviour of native and non-native honey bees, for example in the distance of the mating flight. Furthermore, there are reports of assortative mating within honey bee subspecies driven by a difference in the vertical position of drones [36].

5.4 Halt of imports

Imports of invasive honey bee subspecies continue in Ireland in the face of strong opposition by all beekeeper organisations on the Island. Evidence of introgression is reportedly more apparent both via beekeeper observation and via molecular studies [69]. Legislation to ban imports of honey bees is being considered by the Irish parliament, which is a lengthy process. The Protection of the Native Irish Honey Bee Bill was introduced to the Seanad in October

2022 and was passed through all stages of that house in November 2024. It has recently entered into the Dail business but has not yet been progressed there. In the UK, numbers of imports have been much higher with the result that most honey bees in the UK are introgressed [33]. However, there are pockets of pure and nearly pure local populations still in existence [24]. Is it too late to save A. m. mellifera in the UK or Ireland? What might happen if imports were stopped? Would introgression disappear and if so how long might it take?.

The simulation showed that importing for 10 years (giving about 14% introgression at the rate of +0.006/year) and then halting imports for 10 years only slightly decreased the level of introgression (to 13% at the rate of -0.001/year). In addition, there was a large amount of variation in observed levels of introgression between replicates, ranging between about 7% and 22%.

This is in line with the above discussion that the non-native alleles have been largely invisible to natural stabilising selection on phenotypic values for fitness. Hence, while the new imports have been halted, the previously imported non-native alleles have been segregating in the population and the rate of purging these alleles was estimated to be only -0.001/year. At this rate and assuming a linear relationship, it would take about 13,000 years to purge all non-native alleles. These results highlight the difficulty of reversing introgression once it has occurred, especially in species with high dispersal like honey bees [66]. This is consistent with observations in many other species with documented introgression; non-native alleles have been observed to persist for decades even after the hybridising with the non-native species had stopped [26, 71, 59]. While alleles associated with mal-adaptation and lower fitness in Ireland are likely to reduce in frequency quickly under natural selection, artificial maintenance of alleles by beekeepers 'propping up' susceptible colonies will also maintain these negative alleles in the population. On this basis there is an urgency to prevent any further introgression in the Irish population while it is still largely distinct and well adapted

to local conditions. If major disruption occurs it will be nigh on impossible to reverse the damage.

5.5 Limitations

There is a considerable debate and several reports about the impact of importing non-native honey bees on native populations [10, 43, 47, 29]. While these reports provide data points about the impact, much is unknown on how this impact comes about from the genetic perspective. In the following we describe a number of limitations that could have impacted our results in different ways. The number of limitations is extensive due to the lack of detailed knowledge about many genetic and environmental aspects related to native and non-native honey bees. We acknowledge this challenge and while we could test many possible permutations of the model parameter values, the exponential number of permutations makes this very challenging. Nevertheless, we believe that the simulation results give useful guidelines with respect to honey bee imports and a way forward (see next section). Also, we are providing an open-source stochastic model, which can be revised and extended as needed and when new knowledge becomes available.

With respect to quantitative genetics, there is limited knowledge about native and non-native allele (additive) and genotype (dominance) effects, their interactions with different environments (GxE), their interactions with different genetic backgrounds (epistasis or GxG), artificial and natural selection coefficients, the form of selection (directional, stabilising, frequency-dependent, ...), genotype to phenotype map, and environmental or management effects. In the absence of this knowledge, our trait simulation was guided by reports of higher honey yields in *A. m. ligustica* and *A. m. carnica* than in other honey bee subspecies [7, 64] , and of higher survival of native honey bees compared to non-native honey bees due to better adaptation to the environment [47, 43]. While these studies provided useful guidance, we still had to choose a number of quantitative genetics parameters and each of these could impact

the simulation in a different way. We assumed a complex polygenic architecture with only additive effects for honey yield and fitness, published level of heritabilities for honey yield and fitness, and genetic correlation for honey yield, while we varied the genetic correlation for fitness.

We made additional assumptions related to mating and selection. We assumed random mating, either in a non-spatial or spatial context, and did not account for any mating preferences of the subspecies [36]. If we would have assumed the latter, the rate of introgression would decrease. There is also a lack of studies that quantified the form and strength of selective pressure on native and non-native honey bees. We assumed different types of selection in the burn-in (to generate genetic differentiation) and in the evaluation phase. In the latter we assumed that the native population is at the optimal value for fitness and stabilising selection around this value. This choice, combined with the complex polygenic genetic architecture and low heritability for fitness, meant that selection against non-native alleles was very mild. While the form of selection is maybe questionable, the choice of complex polygenic genetic architecture and low heritability for fitness is in line with published literature [49, 37]. The advent of abundant genome-wide genotype data will hopefully enable studies that will provide more clarity about the form and strength of selection.

Our simulations showed a high variability across replicates. This is not a limitation, but it shows that there is a huge amount of randomness in introgression outcomes, driven by the randomness of genetic inheritance, coupled with selection on phenotypes that are a noisy representation of genetic value of individuals. Future work could focus on increasing the number of replicates to better quantify this amount of variability. The SIMplyBee simulator [51] is still being optimised to work with a large number of colonies and the computational resource was the main factor that determined how many replicates we ran.

Finally, to reduce the computational burden of the simulation, we simulated only 500 colonies, which is only a fraction of the Irish population. This small population size might

have exacerbated the intogression as well as weakened the selection pressure/function on the imported individuals (because drift was sizeable). However, if the imports are mainly limited to few points of entry and there is no additional migration of colonies, our simulation represents the likely situation around these points of entry. To reduce the computational burden even further, we simulated only 10 workers per colony compared to the much larger number of workers found in real colonies (around 50,000). However, we do not believe this has impacted the results significantly because we measured introgression and mean genetic value of the traits on the queens.

5.6 A way forward?

The simulation highlights the likely impact of introgression of non-native alleles into a native honey bee population due to import of queens. Assuming that fitness is a complex polygenic trait with low heritability and that there are significant differences in honey yield between native and non-native populations with medium-to-high genetic correlation, we observed that introgression will happen readily and that it will rapidly spread from the point of entry, increasing genetic value for honey yield and decreasing genetic value for fitness. Based on the documented reports and estimated level of introgression in Ireland [41, 69], all this has already likely happened in the Irish population and the pertinent question is "What is the way forward?". While the short-term increase in honey yield is driving the interest of the importers, it is generating a long-term decrease in fitness for the whole of population. Some beekeepers will benefit from this import even in the long-term, assuming they can provide management (improved environment) that will counteract the negative fitness effects. Other beekeepers will likely not benefit in the long-term, particularly if they use beekeeping practices that have been developed alongside the native population adapted to the local environment. The amount of introgression in the long-term will depend on a complex interplay between the benefits of higher honey yield and costs of improved management, between

beekeepers that want imports and those that don't, their selective breeding actions, and the strength and direction of natural selection. Climate change will likely be a significant factor in this process. As shown in the results, even a small percentage of imports leads to an introgression of non-native alleles. The effect of these non-native alleles in a new environment is largely unknown and will likely depend on the origin of import (importing from similar or different environments). These effects could be negative, neutral, or positive. If negative, the introgressed individuals will show lower preference and/or fitness, which is a phenomena known as outbreeding depression, often observed when individuals from genetically distinct populations or environments interbreed [57]. However, this new source of genetic variation could also be a source for selection to operate on and an opportunity to purge undesired alleles and increase frequency of desirable alleles. This type of a problem is well known also in other species of livestock and crops [e.g. 46, 20] and further research is needed to find optimal ways to leverage this genetic diversity to further improve introgressed/admixed populations.

To avoid loosing the identity of the native populations and to prevent the risk of loosing its native adaptations due to imports, the best course of action should be "conservation by use". Commercial beekeepers import non-native honey bee to improve short-term profitability. However, studies have reported that native honey bees in Ireland and England have the capacity to forage, survive, and mate in lower temperatures [33]. These adaptations have evolved over many years and could be impacted or even lost due to introgression. By establishing selective breeding programmes aimed at enhancing honey yield of the native populations (or even now introgressed/admixed populations) under local environmental conditions, beekeepers could improve commercial viability while preserving the past genetic adaptations and the identity of the native honey bee populations. In fact, countries known for exporting their subspecies globally have established selective breeding programmes. For example, Italy has been selectively breeding for higher productivity and docility for decades

[4]. Other European countries such as Germany, Poland, and Switzerland have also been successful at breeding for increased brood viability or hygienic behavior [68]. Therefore, adopting these selective breeding concepts in Ireland is a way forwards to develop a viable native honey bee population, which balances higher honey yields and other desired traits, while preserving the invaluable local adaptations.

6 Conclusion

In summary, our results demonstrate that although importing non-native honey bees can increased production traits, imports will also increase introgression of the non-native alleles, which could be associated with a decline in fitness and hence the native adaptation to the environment. Moreover, the spread of non-native alleles can be fast and substantial with continued imports and the introgression can not be reversed quickly, even when fully halting imports, meaning that non-native alleles will segregate in the introgressed population for a long time. These findings highlight the importance of strategies to manage genetically distinct subspecies, like *A. m. mellifera* in Ireland and other North European countries. While imposing strict import regulations seems an obvious strategy, it is important to understand and address the reasons for importing non-native honey bees. A comprehensive approach would be (1) to enhance economically important traits of the native honey bees via selective breeding, (2) to raise awareness of the expected level of production of different genotypes in different environments, and (3) to communicate about the risk of disease introduction and short- versus long-term effects of introgressing non-native alleles. In this way, beekeepers can achieve commercial viability without the risk of losing advantageous native adaptations that have evolved over many generations. To support such an approach it is important to expand population and quantitative genetics research of honey bee populations.

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8 Supplementary material

Link to the GitHub repository with code for the simulations:

https://github.com/HighlanderLab/idecarlos_honeybee_import.

Table 3: ANOVA p-values for the rate of introgression, introgression at year 20, honey yield at year 20, and fitness at year 20 on the percentage of imports scenarios and the fitness genetic correlation scenarios

	Rate of introgression	Introgression at year 20	Honey yield at year 20	Fitness at year 20
Percentage of imports	$p < 2e\text{-}16$	$p < 2e\text{-}16$	7.69e-15	3.99e-05
Fitness genetic correlations	0.4760	0.2210	0.2260	0.0093

Table 4: Post-hoc Tukey pairwise comparison p-values between the different percentage of imports scenarios and the different fitness genetic correlation scenarios for the rate of introgression, introgression at year 20, honey yield at year 20, and fitness at year 20.

	Scenario	Rate of introgression	Introgression at year 20	Honey yield at year 20	Fitness at year 20
Percentage of imports	Control - 1.5%	0.0000	0.0000	0.2780	0.9858
	Control - 4%	0.0000	0.0000	0.0001	0.2006
	Control - 10%	0.0000	0.0000	0.0000	0.0000
	4% - 1.5%	0.0000	0.0000	0.0267	0.3571
	4% - 10%	0.0000	0.0000	0.0000	0.0108
	10% - 1.5%	0.0000	0.0000	0.0000	0.0000
Genetic correlations	0.25 - -0.25	0.9230	0.9933	0.4100	0.8576
	0.75 - 0.25	0.9095	0.7120	0.8172	0.0180
	0.25 - -0.75	0.9994	0.8660	0.5053	0.7157
	0.75 - -0.75	0.4983	0.1640	0.8903	0.0086
	0.75 - -0.25	0.5715	0.5470	0.9032	0.1216

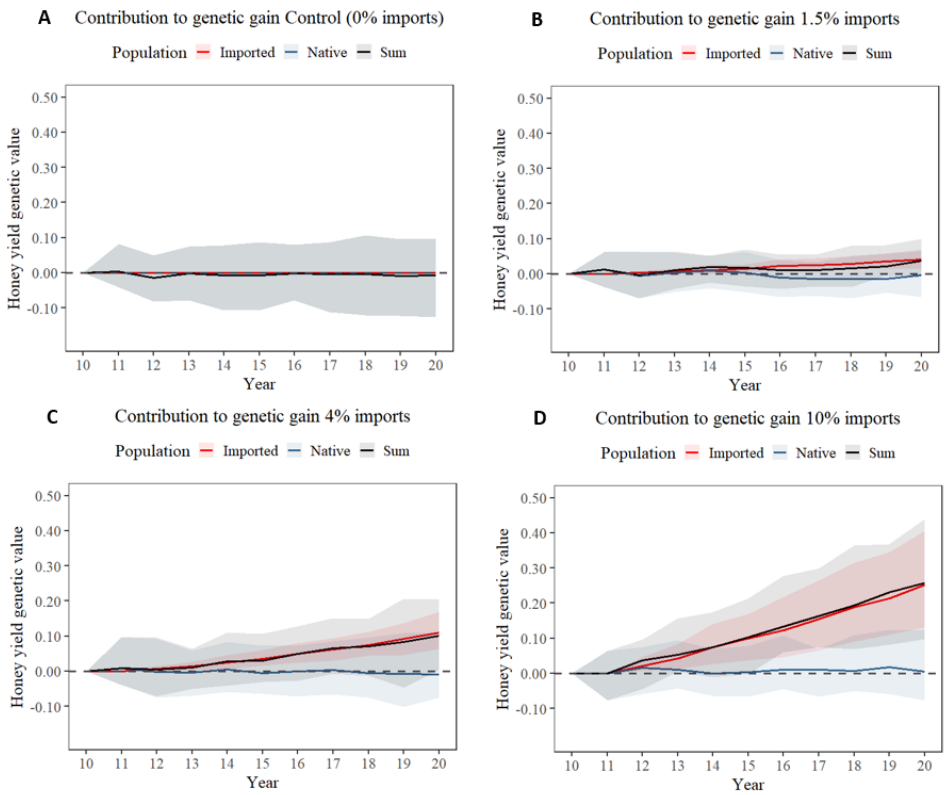

Figure 9: Contribution to the mean genetic value for honey yield from year 10 to year 20, comparing the imported non-native (red line) and native contributions (blue line) to the overall mean genetic value for the Irish population (black line). This is represented across varying percentages of imports: A) Control (0%), B) 1.5%, C) 4%, and D) 10%. Shaded regions indicated the 95% quantiles across replicates.

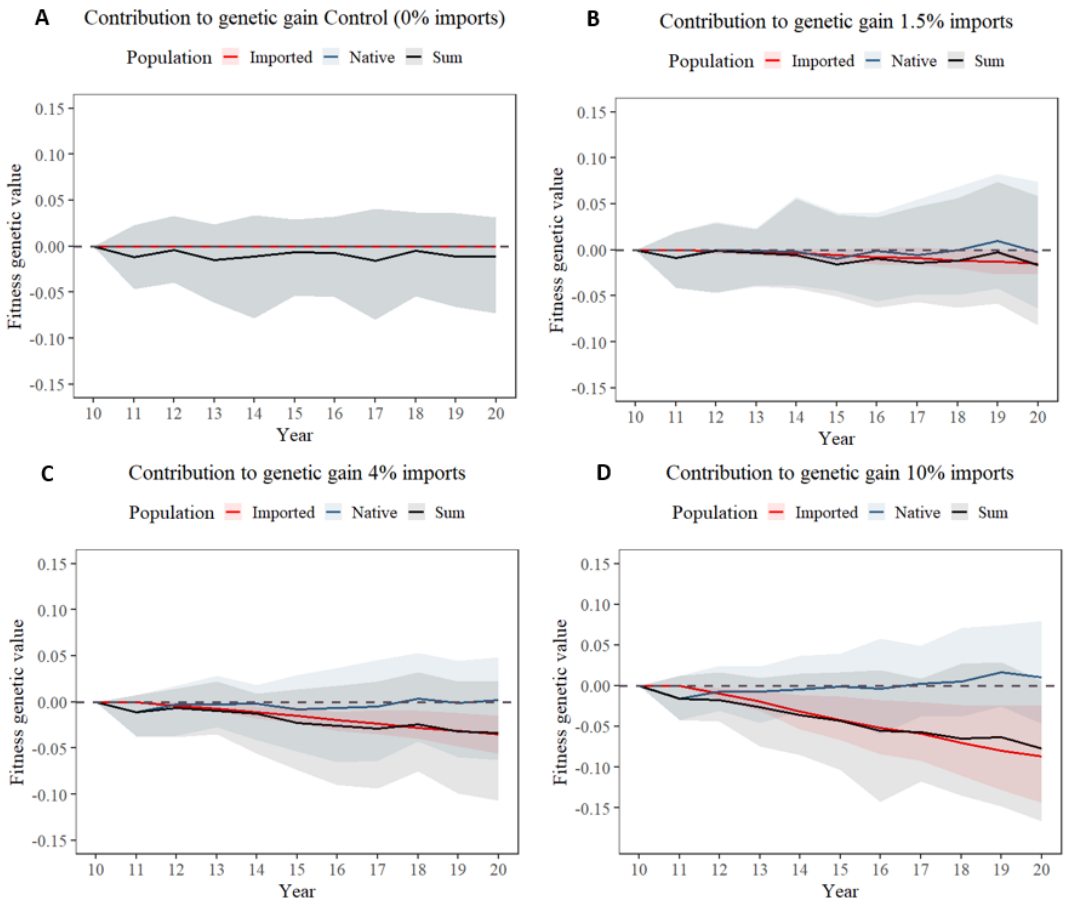

Figure 10: Contribution to the mean genetic value for fitness from year 10 to year 20, comparing the imported non-native (red line) and native contributions (blue line) to the overall mean genetic value for the Irish population (black line). This is represented across varying percentages of imports: A) Control (0%), B) 1.5%, C) 4%, and D) 10%. Shaded regions indicated the 95% quantiles across replicates.

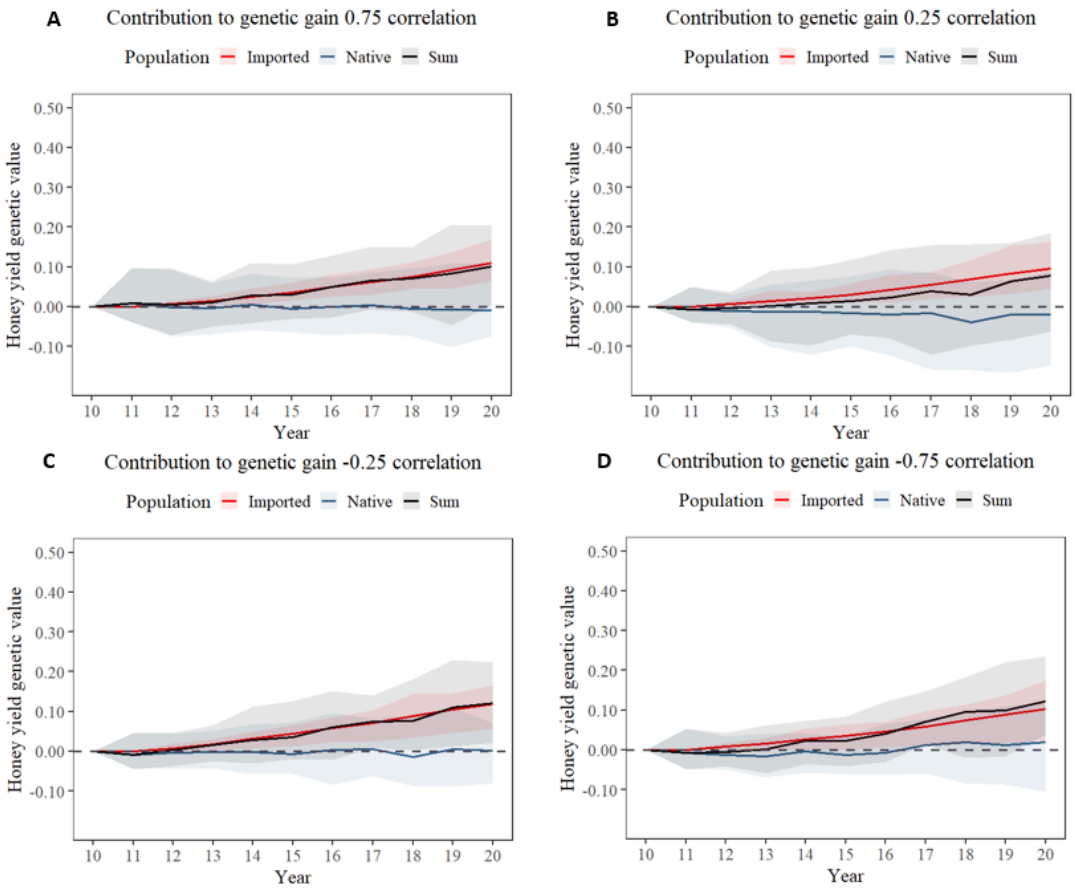

Figure 11: Contribution to the mean genetic value for honey yield from year 10 to year 20, comparing the imported non-native (red line) and native contributions (blue line) to the overall mean genetic value for the Irish population (black line). This is represented across varying genetic correlations between fitness traits: A) +0.75, B) +0.25, C) -0.25, and D) -0.75. Shaded regions indicated the 95% quantiles across replicates.

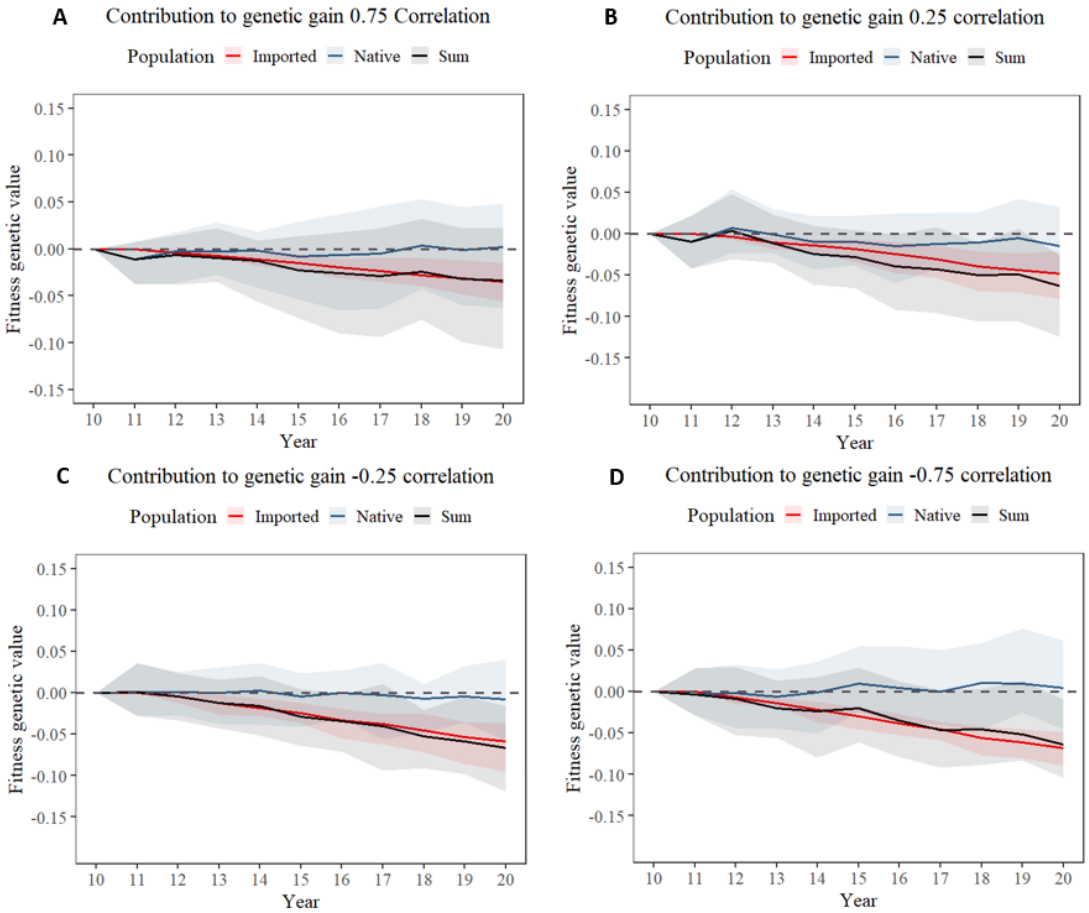

Figure 12: Contribution to the mean genetic value for fitness from year 10 to year 20, comparing the imported non-native (red line) and native contributions (blue line) to the overall mean genetic value for the Irish population (black line). This is represented across varying genetic correlations between fitness traits: A) +0.75, B) +0.25, C) -0.25, and D) -0.75. Shaded regions indicated the 95% quantiles across replicates.

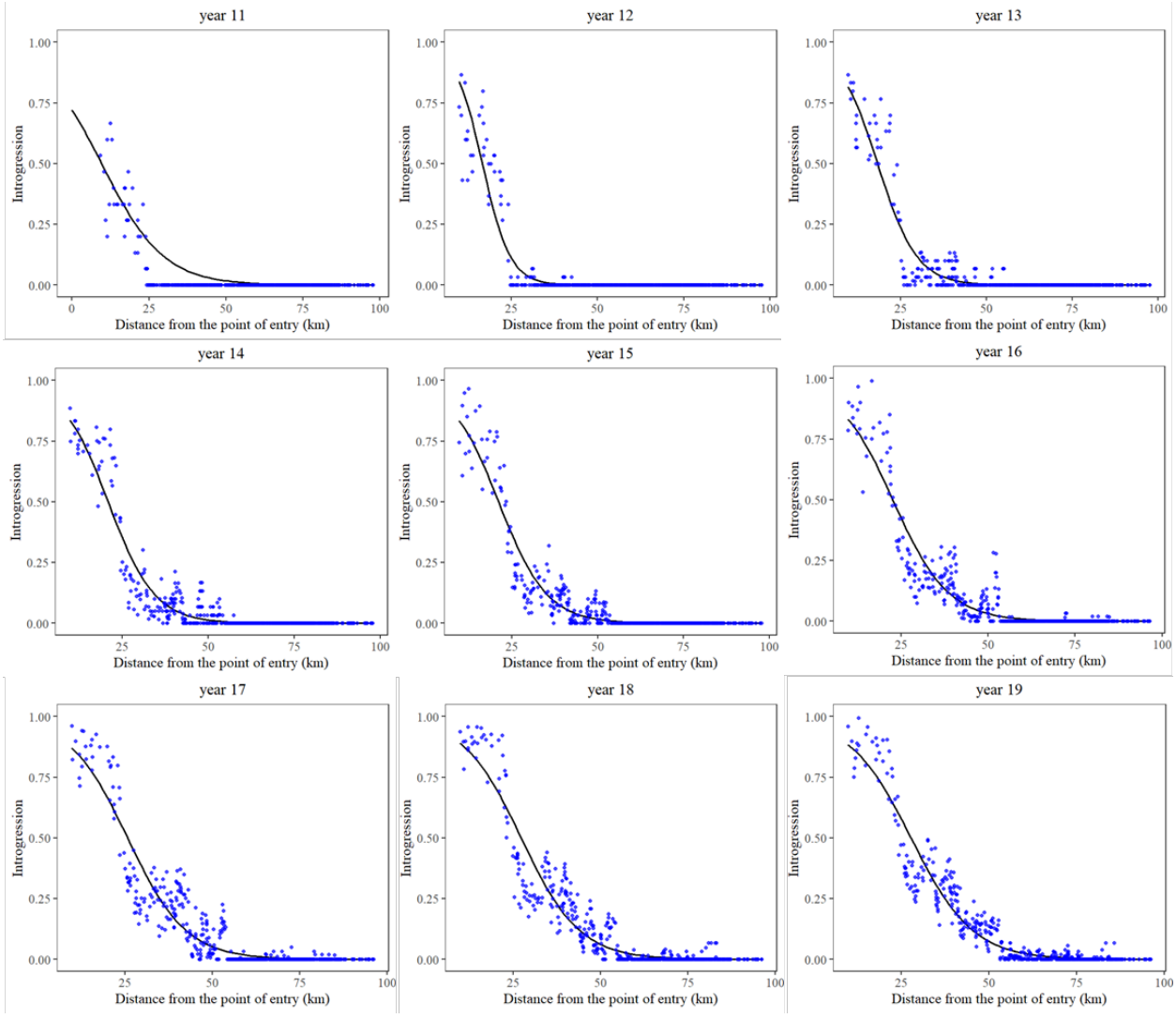

Figure 13: Scatter plot of the introgression levels in honey bee colonies across the distance from the central point of entry on the first (year 11) to the 9th (year 19) year of importation. Each blue dot corresponds to a honey bee colony, with the y-axis indicating the level of introgression and the x-axis representing the distance from the point of entry, which is marked at 0 km. The black line indicates the fitted cline to the scatter plot.

Table 5: Matrix of the p-values of the Post-hoc Tukey pairwise comparisons between the c values (center of the cline) in every year of the importation period.

	11	12	13	14	15	16	17	18	19	20
11	-	0.9368	0.2996	0.0305	0.0804	0.0015	0.0031	0.0001	0.9841	0.0000
12	0.9368	-	0.2996	0.0305	0.0804	0.0015	0.0031	0.0001	0.9841	0.0000
13	0.2996	0.2996	-	0.3208	0.6913	0.0342	0.0181	0.0004	0.0734	0.0001
14	0.0305	0.0305	0.3208	-	0.0194	0.0595	0.1534	0.9954	0.9465	0.0000
15	0.0804	0.0804	0.6913	0.0194	-	0.1843	0.1116	0.0103	0.0000	0.0033
16	0.0015	0.0015	0.0342	0.0595	0.1843	-	0.2411	0.9204	0.9939	0.0000
17	0.0031	0.0031	0.0181	0.1534	0.1116	0.2411	-	0.5094	0.0000	0.0000
18	0.0001	0.0001	0.0004	0.9954	0.0103	0.9204	0.5094	-	0.9999	0.9999
19	0.9841	0.9841	0.0734	0.9465	0.0000	0.9939	0.0000	0.9999	-	0.9999
20	0.0000	0.0000	0.0000	0.0000	0.0033	0.0000	0.0000	0.9999	0.99996	-

9 References

[1] M.C. Arias and W.S. Sheppard. Molecular phylogenetics of honey bee subspecies (apis melliferal.) inferred from mitochondrial dna sequence. *Molecular Phylogenetics and Evolution*, 5(3):557–566, 1996. ISSN 1055-7903. doi: https://doi.org/10.1006/mpev.1996.0050. URL https://www.sciencedirect.com/science/article/pii/S1055790396900500.

[2] J Bančič, P Greenspoon, C R Gaynor, and G Gorjanc. Plant breeding simulations with AlphaSimR. *Crop Science*, 65(1), 2024. doi: 10.1002/csc2.21312. URL http://dx.doi.org/10.1002/csc2.21312.

[3] J. Bančič, B. Ovenden, G. Gorjanc, and D. J. Tolhurst. Genomic selection for genotype performance and stability using information on multiple traits and multiple environments. *TAG. Theoretical and applied genetics. Theoretische und angewandte Genetik*, 136(5):104, April 2023. ISSN 1432-2242. doi: 10.1007/s00122-023-04305-1.

[4] R Bar-Cohen, G Alpern, and R Bar-Anan. Progeny testing and selecting italian queens

for brood area and honey production. *Apidologie*, 9(2):95–100, 1978.

[5] Matthias A. Becher, Volker Grimm, Pernille Thorbek, Juliane Horn, Peter J. Kennedy, and Juliet L. Osborne. BEEHAVE: a systems model of honeybee colony dynamics and foraging to explore multifactorial causes of colony failure. *Journal of Applied Ecology*, 51(2):470–482, 2014. doi: 10.1111/1365-2664.12222. URL https://onlinelibrary.wiley.com/doi/10.1111/1365-2664.12222.

[6] M.M. Benjamin, P. Cheyenne, L. Quinn, L.P. Daniel, B. Yaniv, and S. Molly. The genomic consequences of hybridization . *eLife*, 10:e69016, 2021. URL https://doi.org/10.7554/eLife.69016.

[7] Richard Bernstein, Andreas Hoppe, and Kaspar Bienefeld. A new method for estimating breeding values in honey bees, applied to a local population of the dark european honey bee (Apis mellifera mellifera) in northern germany. *Animals*, 10(11):2158, 2020. doi: 10.3390/ani10112158. URL https://www.ncbi.nlm.nih.gov/pmc/articles/PMC7694995/.

[8] Richard Bernstein, Manuel Du, Andreas Hoppe, and Kaspar Bienefeld. Simulation studies to optimize genomic selection in honey bees. *Genetics Selection Evolution*, 53(1), 2021. doi: 10.1186/s12711-021-00654-x. URL http://dx.doi.org/10.1186/s12711-021-00654-x.

[9] Robert Brodschneider, Alison Gray, Noureddine Adjlane, Alexis Ballis, Valters Brusbardis, Jean-Daniel Charrière, Robert Chlebo, Mary F Coffey, Bjørn Dahle, Dirk C de Graaf, Marica Maja Dražić, Garth Evans, Mariia Fedoriak, Ivan Forsythe, Aleš Gregorc, Urszula Grzęda, Amots Hetzroni, Lassi Kauko, Preben Kristiansen, Maritta Martikkala, Raquel Martín-Hernández, Carlos Aurelio Medina-Flores, Franco Mutinelli, Aivar Raudmets, Vladimir A Ryzhikov, Noa Simon-Delso, Jevrosima Stevanovic, Aleksandar Uzunov, Flemming Vejsnæs, Saskia Wöhl, Marion Zammit-

Mangion, and Jiří Danihlík. Multi-country loss rates of honey bee colonies during winter 2016/2017 from the COLOSS survey. *Journal of Apicultural Research*, 57(3): 452–457, May 2018. ISSN 0021-8839. doi: 10.1080/00218839.2018.1460911. URL https://doi.org/10.1080/00218839.2018.1460911. Publisher: Taylor & Francis _eprint: https://doi.org/10.1080/00218839.2018.1460911.

[10] Ralph Büchler, Cecilia Costa, Fani Hatjina, Sreten Andonov, Marina D Meixner, Yves Le Conte, Aleksandar Uzunov, Stefan Berg, Malgorzata Bienkowska, Maria Bouga, Maja Drazic, Winfried Dyrba, Per Kryger, Beata Panasiuk, Hermann Pechhacker, Plamen Petrov, Nikola Kezić, Seppo Korpela, and Jerzy Wilde. The influence of genetic origin and its interaction with environmental effects on the survival of Apis mellifera L. colonies in Europe. *Journal of Apicultural Research*, 53(2):205–214, January 2014. ISSN 0021-8839. doi: 10.3896/IBRA.1.53.2.03. URL https://doi.org/10.3896/IBRA.1.53.2.03. Publisher: Taylor & Francis _eprint: https://doi.org/10.3896/IBRA.1.53.2.03.

[11] Madeline H. Carpenter and Brock A. Harpur. Genetic past, present, and future of the honey bee (apis mellifera) in the united states of america. *Apidologie*, 52:63—-79, 2021. URL https://doi.org/10.1007/s13592-020-00836-4.

[12] Roselle Chapman. The influence of sociality on the conservation biology of social insects. *Ecology Letters*, January 2001. URL https://www.academia.edu/8916932/The_influence_of_sociality_on_the_conservation_biology_of_social_insects.

[13] Elizabeth T. Chevy, Jiseon Min, Victoria Caudill, Samuel E. Champer, Benjamin C. Haller, Clara T. Rehmann, Chris C. R. Smith, Silas Tittes, Philipp W. Messer, Andrew D. Kern, Sohini Ramachandran, and Peter L. Ralph. Population genetics meets ecology: a guide to individual-based simulations in continuous landscapes. *bioRxiv*, 2024. doi: 10.1101/2024.07.24.604988. URL http://dx.doi.org/10.1101/2024.07.24.604988.

[14] P. De la Rua, R. Jaffé, R. Dall'Olio, I. Muñoz, and J. Serrano. Biodiversity, conservation and current threats to European honeybees . *Molecular Ecology*, 40(3):263–284, 2009. URL https://doi.org/10.1051/apido/2009027.

[15] Department of Agriculture, Food and the Marine. Beekeepers census 2019. https://assets.gov.ie/130944/94accd3e-aa87-4de6-9ce0-ddce1013c5c6.pdf, 2019. Government of Ireland.

[16] Elizabeth P. Derryberry, Graham E. Derryberry, James M. Maley, and Robb T. Brumfield. hzar: hybrid zone analysis using an r software package. *Molecular Ecology Resources*, 14(3):652–663, 2014. doi: https://doi.org/10.1111/1755-0998.12209. URL https://onlinelibrary.wiley.com/doi/abs/10.1111/1755-0998.12209.

[17] Kathleen A Dogantzis and Amro Zayed. Recent advances in population and quantitative genomics of honey bees. *Current Opinion in Insect Science*, 31:93–98, 2019. ISSN 2214-5745. doi: https://doi.org/10.1016/j.cois.2018.11.010. URL https://www.sciencedirect.com/science/article/pii/S2214574518300968. Insect genomics • Development and regulation.

[18] J.S. Ellis, G. Soland-Reckeweg, V.G. Buswell, G.J. Hulm, A. Brown, and M.E. Knight. Introgression in native populations of Apis mellifera mellifera L: implications for conservation . *J Insect Conserv*, 22:377–390, 2018. URL https://doi.org/10.1007/s10841-018-0067-7.

[19] R C Gaynor, G Gorjanc, and J M Hickey. AlphaSimR: An R-package for breeding program simulations. *G3: Genes/Genomes/Genetics*, 11, 2021. doi: 10.1093/g3journal/jkaa017.

[20] G Gorjanc, J Jenko, S J Hearne, and J M Hickey. Initiating maize pre-breeding programs

using genomic selection to harness polygenic variation from landrace populations. *BMC Genomics*, 17(1):1–15, 2016. doi: 10.1186/s12864-015-2345-z.

[21] Alison Gray, Noureddine Adjlane, Alireza Arab, Alexis Ballis, Valters Brusbardis, Adrian Bugeja Douglas, Luis Cadahía, Jean-Daniel Charrière, Robert Chlebo, Mary F. Coffey, Bram Cornelissen, Cristina Amaro da Costa, Ellen Danneels, Jiří Danihlík, Constantin Dobrescu, Garth Evans, Mariia Fedoriak, Ivan Forsythe, Aleš Gregorc, Iliyana Ilieva Arakelyan, Jes Johannesen, Lassi Kauko, Preben Kristiansen, Maritta Martikkala, Raquel Martín-Hernández, Ewa Mazur, Carlos Aurelio Medina-Flores, Franco Mutinelli, Eslam M. Omar, Solenn Patalano, Aivar Raudmets, Gilles San Martin, Victoria Soroker, Philip Stahlmann-Brown, Jevrosima Stevanovic, Aleksandar Uzunov, Flemming Vejsnaes, Anthony Williams, and Robert Brodschneider. Honey bee colony loss rates in 37 countries using the COLOSS survey for winter 2019–2020: the combined effects of operation size, migration and queen replacement. *Journal of Apicultural Research*, 62 (2):204–210, March 2023. ISSN 0021-8839. doi: 10.1080/00218839.2022.2113329. URL https://doi.org/10.1080/00218839.2022.2113329. Publisher: Taylor & Francis _eprint: https://doi.org/10.1080/00218839.2022.2113329.

[22] Matthieu Guichard, Florence Phocas, Markus Neuditschko, Benjamin Basso, and Benjamin Dainat. An Overview of Selection Concepts Applied to Honey Bees. *Bee World*, 100(1):2–8, January 2023. ISSN 0005-772X. doi: 10.1080/0005772X.2022.2147702. URL https://doi.org/10.1080/0005772X.2022.2147702. Publisher: Taylor & Francis _eprint: https://doi.org/10.1080/0005772X.2022.2147702.

[23] J. Hassett, K.A. Browne, G.P. McCormack, E. Moore, Native Irish Honey Bee Society, G. Soland, and G.M. Michael. A significant pure population of the dark European honey bee Apis mellifera mellifera remains in Ireland. *Journal of Apicultural Research*, 57(3): 337–350, 2018. URL https://doi.org/10.1080/00218839.2018.1433949.

[24] Jack Hassett, Keith A Browne, Grace P McCormack, Elizabeth Moore, Native Irish Honey Bee Society, Gabrielle Soland, and Michael Geary. A significant pure population of the dark European honey bee (Apis mellifera mellifera) remains in Ireland. *Journal of Apicultural Research*, 57(3):337–350, May 2018. ISSN 0021-8839. doi: 10.1080/00218839.2018.1433949. URL https://doi.org/10.1080/00218839.2018.1433949. Publisher: Taylor & Francis _eprint: https://doi.org/10.1080/00218839.2018.1433949.

[25] Fani Hatjina, Cecilia Costa, Büchler Ralph, Aleksandar Uzunov, Maja Dražić, Janja Filipi, Leonidas Charistos, Ruottinen L., Sreten Andonov, Marina Meixner, Malgorzata Bienkowska, Gerula D., Beata Panasiuk, Yves Le Conte, Jerzy Wilde, Stefan Berg, Maria Bouga, Winfried Dyrba, Hrisula Kiprijanovska, and Bul Aleksandar. Population dynamics of European honey bee genotypes under different environmental conditions. *Journal of Apicultural Research*, 53:233–247, May 2014. doi: 10.3896/IBRA.1.53.2.05.

[26] Philip W. Hedrick. Conservation Genetics and North American Bison (Bison bison). *Journal of Heredity*, 100(4):411–420, July 2009. ISSN 0022-1503. doi: 10.1093/jhered/esp024. URL https://doi.org/10.1093/jhered/esp024.

[27] D. Henriques, K.A. Browne, M.W. Barnett, and et al. High sample throughput genotyping for estimating C-lineage introgression in the dark honeybee: an accurate and cost-effective SNP-based tool. *Sci Rep*, 8:8552, 2018. URL https://doi.org/10.1038/s41598-018-26932-1.

[28] D. Henriques, A.R. Lopes, R. Ferrari, and et al. Can introgression in M-lineage honey bees be detected by abdominal colour patterns? . *Apidologie*, 51:583–593, 2020. URL https://doi.org/10.1007/s13592-020-00744-7.

[29] Dora Henriques, Keith A. Browne, Mark W. Barnett, Melanie Parejo, Per Kryger, Tom C. Freeman, Irene Muñoz, Lionel Garnery, Fiona Highet, J. Spencer Jonhston,

Grace P. McCormack, and M. Alice Pinto. High sample throughput genotyping for estimating C-lineage introgression in the dark honeybee: an accurate and cost-effective SNP-based tool. *Scientific Reports*, 8(1):8552, June 2018. ISSN 2045-2322. doi: 10.1038/s41598-018-26932-1. URL https://www.nature.com/articles/s41598-018-26932-1. Number: 1 Publisher: Nature Publishing Group.

[30] A. Hoppe, M. Du, R. Bernstein, F.K. Tiesler, M. Kärcher, and K. Bienefeld. Substantial Genetic Progress in the International Apis mellifera carnica Population Since the Implementation of Genetic Evaluation . *Insects*, 11(11):768, 2020. URL https://doi.org/10.3390/insects11110768.

[31] R. A. Ilyasov, M. L. Lee, J. I. Takahashi, H. W. Kwon, and A. G. Nikolenko. A revision of subspecies structure of western honey bee Apis mellifera. *Saudi J Biol Sci*, 27(12):3615–3621, 2020. URL https://doi.org/10.1016/j.sjbs.2020.08.001.

[32] A.B. Jensen, K.A. Palmer, J.J. Boomsma, and B.V. Pedersen. Varying degrees of Apis mellifera ligustica introgression in protected populations of the black honeybee ,Apis mellifera mellifera, in northwest Europe . *Molecular Ecology*, 14(1):93–10, 2005a. URL https://doi.org/10.1111/j.1365-294X.2004.02399.x.

[33] Annette B. Jensen, Kellie A. Palmer, Nicolas Chaline, Nigel E. Raine, Adam Tofilski, Stephen J. Martin, Bo V. Pedersen, Jacobus J. Boomsma, and Francis L.W. Ratnieks. Quantifying honey bee mating range and isolation in semi-isolated valleysby DNA microsatellite paternity analysis. *Conservation Genetics*, 6(4):527–537, July 2005. ISSN 1572-9737. doi: 10.1007/s10592-005-9007-7. URL https://doi.org/10.1007/s10592-005-9007-7.

[34] Ben Jones and Nigel Semmence. Imported honey bees: Risks and mitigation. *BBKA*

News Incorporating The British Bee Journal, 68:295–296, September 2021. Published by Fera Science Ltd and APHA.

[35] Tristan Kistler, Benjamin Basso, and Florence Phocas. A simulation study of a honeybee breeding scheme accounting for polyandry, direct and maternal effects on colony performance. *Genetics Selection Evolution*, 53(1), 2021. doi: 10.1186/s12711-021-00665-8. URL http://dx.doi.org/10.1186/s12711-021-00665-8.

[36] G. Koeniger, N. Koeniger, H. Pechhacker, F. Ruttner, and S. Berg. Assortative mating in a mixed population of European Honeybees,Apis mellifera ligustica andApis mellifera carnica. *Insectes Sociaux*, 36(2):129–138, June 1989. ISSN 1420-9098. doi: 10.1007/BF02225908. URL https://doi.org/10.1007/BF02225908.

[37] Nikolaus Koeniger, Gudrun Koeniger, and Heinrich Pechhacker. The nearer the better? drones (Apis mellifera) prefer nearer drone congregation areas. *Insectes Sociaux*, 46: 323–327, 1999. doi: 10.1007/s000400050159. URL https://pubmed.ncbi.nlm.nih.gov/10639142/.

[38] L. E. Kruuk, T. H. Clutton-Brock, J. Slate, J. M. Pemberton, S. Brotherstone, and F. E. Guinness. Heritability of fitness in a wild mammal population. *Proceedings of the National Academy of Sciences of the United States of America*, 97(2):698–703, January 2000. ISSN 0027-8424. doi: 10.1073/pnas.97.2.698.

[39] H. R. Mattila and G. W. Otis. Influence of Pollen Diet in Spring on Development of Honey Bee (Hymenoptera: Apidae) Colonies. *Journal of Economic Entomology*, 99(3):604–613, June 2006. ISSN 0022-0493. doi: 10.1093/jee/99.3.604. URL https://doi.org/10.1093/jee/99.3.604.

[40] Ségolène Maucourt, Frédéric Fortin, Claude Robert, and Pierre Giovenazzo. Genetic

Parameters of Honey Bee Colonies Traits in a Canadian Selection Program. *Insects*, 11 (9):587, September 2020. ISSN 2075-4450. doi: 10.3390/insects11090587.

[41] M. McCann and G.P. McCormack. Increased levels of introgression evident in Irish honey bees. *Journal of Apicultural Research*, 63(1):205–207, 2024. URL https://doi.org/10.1080/00218839.2023.2262872.

[42] M.L. McKinney and J.L. Lockwood. Biotic homogenization: a few winners replacing many losers in the next mass extinction. *Trends in ecology and evolution*, 14(11):450–453, 1999. URL https://doi.org/10.1016/S0169-5347(99)01679-1.

[43] Marina D Meixner, Per Kryger, and Cecilia Costa. Effects of genotype, environment, and their interactions on honey bee health in europe. *Current Opinion in Insect Science*, 10: 177–184, 2015. ISSN 2214-5745. doi: https://doi.org/10.1016/j.cois.2015.05.010. URL https://www.sciencedirect.com/science/article/pii/S2214574515000899. Social Insects * Vectors and Medical and Veterinary Entomology.

[44] G. Minozzi, B. Lazzari, M.G. De Iorio, C. Costa, E. Carpana, P. Crepaldi, R. Rizzi, E. Facchini, G. Gandini, A. Stella, and G. Pagnacco. Whole-Genome Sequence Analysis of Italian Honeybees Apis mellifera . *Animals (Basel)*, 11(5):1311, 2021. URL https://doi.org/10.3390/ani11051311.

[45] J. Momeni, M. Parejo, R.O. Nielsen, and et al. Authoritative subspecies diagnosis tool for European honey bees based on ancestry informative SNPs. *BMC Genomics*, 22(1): 101, 2021. URL https://doi.org/10.1186/s12864-021-07379-7.

[46] Okeyo Mwai, Olivier Hanotte, Young-Jun Kwon, and Seoae Cho. African indigenous cattle: Unique genetic resources in a rapidly changing world. *Asian-Australasian Journal of Animal Sciences*, 28(7):911—921, 2015. doi: 10.5713/ajas.15.0002r. URL http://dx.doi.org/10.5713/ajas.15.0002R.

[47] Ndoumbe Ndiaye. Analysis of iberian honey bee (apis mellifera iberiensis) colony phenotypes across two different origins in portugal: Searching for evidence of genotype by environment interactions. Mestrado em recursos florestais, Instituto Politécnico de Bragança, 2017. URL https://bibliotecadigital.ipb.pt/handle/10198/14431.

[48] M. Niklasson, E. Svensson, S. Leidenberger, and et al. Free-living colonies of native honey bees (*Apis mellifera mellifera*) in 19th and early 20th century Sweden. *J Insect Conserv*, 2023. URL https://doi.org/10.1007/s10841-023-00541-4.

[49] Magnus Niklasson, Thomas Purkiss, and Robert Brodschneider. Evaluating the conservation status of the dark european honeybee (Apis mellifera mellifera) using community science data from the uk and ireland. *Bee World*, 99(4):134–140, 2022. doi: 10.1080/0005772X.2022.2147702. URL https://www.tandfonline.com/doi/full/10.1080/0005772X.2022.2147702.

[50] Jana Obšteter, Justin Holl, John M. Hickey, and Gregor Gorjanc. AlphaPart—R implementation of the method for partitioning genetic trends. *Genetics Selection Evolution*, 53(1):30, March 2021. ISSN 1297-9686. doi: 10.1186/s12711-021-00600-x. URL https://doi.org/10.1186/s12711-021-00600-x.

[51] Jana Obšteter, Laura K. Strachan, Jernej Bubnič, Janez Prešern, and Gregor Gorjanc. SIMplyBee: an R package to simulate honeybee populations and breeding programs. *Genetics, Selection, Evolution : GSE*, 55:31, May 2023. ISSN 0999-193X. doi: 10.1186/s12711-023-00798-y. URL https://www.ncbi.nlm.nih.gov/pmc/articles/PMC10169377/.

[52] M.A. Pinto, D. Henriques, J. Chávez-Galarza, P. Kryger, L. Garnery, and et al. Genetic integrity of the Dark European honey bee Apis mellifera mellifera from protected pop-

ulations: a genome-wide assessment using SNPs and mtDNA sequence data. *Journal of Apicultural Research*, 53:269–278, 2014. URL https://doi.org/10.3896/IBRA.1.53.2.08.

[53] Manuel Plate, Richard Bernstein, Andreas Hoppe, and Kaspar Bienefeld. Comparison of infinitesimal and finite locus models for long-term breeding simulations with direct and maternal effects at the example of honeybees. *PLOS ONE*, 14(3):e0213270, 2019. doi: 10.1371/journal.pone.0213270. URL http://dx.doi.org/10.1371/journal.pone.0213270.

[54] Torsten Pook, Martin Schlather, and Henner Simianer. MoBPS - modular breeding program simulator. *G3 Genes/Genomes/Genetics*, 10(6):1915–1918, 2020. doi: 10.1534/g3.120.401193. URL https://doi.org/10.1534/g3.120.401193.

[55] E. Péntek-Zakar, A. Oleksa, T. Borowik, and S. Kusza. Population structure of honey bees in the Carpathian Basin (Hungary) confirms introgression from surrounding subspecies . *Ecol Evol*, 5(23):5456–5467, 2015. URL https://doi.org/10.1002/ece3.1781.

[56] R Core Team. *R: A Language and Environment for Statistical Computing*. R Foundation for Statistical Computing, Vienna, Austria, 2024. URL https://www.R-project.org/.

[57] Katherine Ralls, Richard Frankham, and Jonathan D. Ballou. Inbreeding and outbreeding. In Simon A Levin, editor, *Encyclopedia of Biodiversity (Second Edition)*, pages 245–252. Academic Press, Waltham, second edition edition, 2013. ISBN 978-0-12-384720-1. doi: https://doi.org/10.1016/B978-0-12-384719-5.00073-3. URL https://www.sciencedirect.com/science/article/pii/B9780123847195000733.

[58] Juliana Rangel, J. Keller, and David Tarpy. The effects of honey bee (Apis mellifera L.) queen reproductive potential on colony growth. *Insectes Sociaux*, 60, February 2013. doi: 10.1007/s00040-012-0267-1.

[59] Judith M. Rhymer and Daniel Simberloff. Extinction by Hybridization and Introgression. *Annual Review of Ecology and Systematics*, 27(1):83–109, 1996. doi:

10.1146/annurev.ecolsys.27.1.83. URL https://doi.org/10.1146/annurev.ecolsys.27.1.83. _eprint: https://doi.org/10.1146/annurev.ecolsys.27.1.83.

[60] L. Ruottinen, B. Peer, J. Kantanen, T. Kristensen, and A. Kettunen. Status and Conservation of the Nordic Brown Bee: Final Report. *Nord Gen*, 15, 2014.

[61] Kevin M. Schultz, Kevin M. Passino, and Thomas D. Seeley. The mechanism of flight guidance in honeybee swarms: subtle guides or streaker bees? *Journal of Experimental Biology*, 211(20):3287–3295, October 2008. ISSN 0022-0949. doi: 10.1242/jeb.018994. URL https://doi.org/10.1242/jeb.018994.

[62] Nicola Seitz, Kirsten S. Traynor, Nathalie Steinhauer, Karen Rennich, Michael E. Wilson, James D. Ellis, Robyn Rose, David R. Tarpy, Ramesh R. Sagili, Dewey M. Caron, Keith S. Delaplane, Juliana Rangel, Kathleen Lee, Kathy Baylis, James T. Wilkes, John A. Skinner, Jeffery S. Pettis, and Dennis vanEngelsdorp. A national survey of managed honey bee 2014–2015 annual colony losses in the USA. *Journal of Apicultural Research*, 54(4):292–304, August 2015. ISSN 0021-8839. doi: 10.1080/00218839.2016.1153294. URL http://www.scopus.com/inward/record.url?scp=84969850493&partnerID=8YFLogxK.

[63] Stephen Smith, Arrigo Moro, and Grace P. McCormack. Exploring a Potential Avenue for Beekeeping in Ireland: Safeguarding Locally Adapted Honeybees for Breeding Varroa-Resistant Lines. *Insects*, 14(10):827, October 2023. ISSN 2075-4450. doi: 10.3390/insects14100827. URL https://www.ncbi.nlm.nih.gov/pmc/articles/PMC10607453/.

[64] Georgina Strachan, Kaspar Bienefeld, and Laurence Guichard. Principles of breeding for resistance to varroa destructor in honey bees. *Biology*, 13(2):206, 2024. doi: 10.3390/biology13020206. URL https://www.ncbi.nlm.nih.gov/pmc/articles/PMC11249005/.

[65] L Strachan, J Bubnič, G Petersen, **G Gorjanc**, and J Obšteter. The principles of expected and realised genetic relatedness among individual honeybees. *bioRxiv*, 2024. doi: 10.1101/2024.05.26.595938. URL https://doi.org/10.1101/2024.05.26.595938.

[66] David A. Tallmon, Gordon Luikart, and Robin S. Waples. The alluring simplicity and complex reality of genetic rescue. *Trends in Ecology & Evolution*, 19(9):489–496, September 2004. ISSN 0169-5347. doi: 10.1016/j.tree.2004.07.003.

[67] D. Seeley Thomas. Honeybee Ecology | Princeton University Press, April 2016. URL https://press.princeton.edu/books/hardcover/9780691639352/honeybee-ecology. ISBN: 9780691639352.

[68] Aleksandar Uzunov, Cecilia Costa, Beata Panasiuk, Marina Meixner, Per Kryger, Fani Hatjina, Maria Bouga, Sreten Andonov, Malgorzata Bienkowska, Yves Le Conte, Jerzy Wilde, Gerula D., Hrisula Kiprijanovska, Janja Filipi, Plamen Petrov, Ruottinen L., Hermann Pechhacker, and Stefan Berg. Swarming, defensive and hygienic behaviour in honey bee colonies of different genetic origin in a pan-european experiment. *Journal of Apicultural Research*, 53:248–260, 05 2014. doi: 10.3896/IBRA.1.53.2.06.

[69] Alexandra Valentine, Arrigo Moro, Ethan Briggs, Brandon Collier, Kenneth Sandoval, Chiara Binetti, Matthew Richardson, David Wragg, Keith Browne, Mark Barnett, and Grace Mccormack. Introgressive hybridisation puts the distinctive population of apis mellifera mellifera in ireland at risk: evidence from a multidisciplinary approach. *Journal of Apicultural Research*, 64:1–15, 10 2024. doi: 10.1080/00218839.2024.2404297.

[70] Andreas Wallberg, Fan Han, Gustaf Wellhagen, Bjørn Dahle, Masakado Kawata, Nizar Haddad, Zilá Luz Paulino Simões, Mike H. Allsopp, Irfan Kandemir, Pilar De la Rúa, Christian W. Pirk, and Matthew T. Webster. A worldwide survey of genome sequence variation provides insight into the evolutionary history of the honeybee Apis mellifera.

Nature Genetics, 46(10):1081–1088, October 2014. ISSN 1546-1718. doi: 10.1038/ng.3077.

[71] Robert K. Wayne and H. Bradley Shaffer. Hybridization and endangered species protection in the molecular era. *Molecular Ecology*, 25(11):2680–2689, 2016. doi: https://doi.org/10.1111/mec.13642. URL https://onlinelibrary.wiley.com/doi/abs/10.1111/mec.13642.