

Inheritance of Resistance to *Acarapis woodi* (Acari: Tarsonemidae) in Crosses Between Selected Resistant Russian and Selected Susceptible U.S. Honey Bees (Hymenoptera: Apidae)

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ABSTRACT The pattern of inheritance of tracheal mite resistance in selected Russian bees was determined in bioassays and in samples from field colonies. Resistant colonies of Russian origin and colonies selected for high susceptibility in the United States were used to generate divergent parental populations. Seven groups of F₁ colonies were produced by crossing queens and drones from these selected resistant Russian and selected susceptible populations. In a series of bioassays with young workers exposed in infested colonies, average mite abundance (female mites per worker) in F₁ colonies was intermediate (1.04 ± 0.13 [mean \pm SE]) and significantly different from that of both resistant Russian (0.74 ± 0.13) and selected susceptible (1.57 ± 0.13) colonies. Colonies representing the three populations were established in two apiaries in July 2005. Colonies surviving with original queens after 10 mo had mite prevalences supporting the findings of the bioassay. All three resistant colonies had undetectable mite levels, whereas prevalences in four F₁ colonies ranged from 0 to 53%, and in 10 susceptible colonies ranged from 0 to 90%. Tracheal mite resistance in Russian bees is likely polygenic, but there may be a number of genes with major dominance interacting with minor genes. Use of selected Russian queens mated with Russian drones or with drones from unknown sources is beneficial for beekeeping in areas with persistent problems with tracheal mite infestation.

KEY WORDS *Apis mellifera*, *Acarapis woodi*, genetic resistance, honey bees, tracheal mites

Parasitism of honey bees, *Apis mellifera* L. (Hymenoptera: Apidae), by tracheal mites, *Acarapis woodi* (Rennie) (Acari: Tarsonemidae), continues to produce problems for beekeepers. Susceptible colonies often are weakened or killed when tracheal mite populations are high (Eischen 1987, Otis and Scott-Dupree 1992, De Guzman 2006). However, strains of bees with genetic resistance to tracheal mites have few or no problems (Milne et al. 1991, Szabo et al. 1991, Rinderer et al. 1993, Danka et al. 1995, Lin et al. 1996). Russian honey bees, imported by and developed by USDA-ARS, have generally high levels of genetic resistance to tracheal mites and seldom reach levels considered harmful (De Guzman et al. 2002, 2006).

Tracheal mite resistance is heritable and responds to selection (Page and Gary 1990, Nasr et al. 2001). Experiences with Buckfast bees in Great Britain showed that hybrid bees maintain field infestations as low as resistant parental colonies (Calvert 1957, Adam 1968). A similar pattern was found in crosses of resistant colonies from Yugoslavia (ARS-Y-C-1) with susceptible colonies (Rinderer et al. 1993). When Buckfast bees imported to North America were crossed with different sources of susceptible colonies, hybrids also had low levels of infestation in the field (Lin et al.

1996) and in bioassays (Lin et al. 1996, Danka and Villa 2000), suggesting partial or incomplete dominance.

Bioassays and field evaluations were conducted to infer the inheritance of tracheal mite resistance in hybrids of selected resistant Russian \times selected susceptible bees. Understanding the genetics of inheritance can guide the efficient release of genetic material and the optimal application of breeding methods in commercial queen production. In addition, understanding the pattern of inheritance of the trait can be used to design experiments to develop potential molecular markers for marker assisted selection.

Materials and Methods

Seven groups of F₁ colonies were produced from crosses of selected highly resistant Russian and selected highly susceptible colonies from the United States. Bioassays (Gary and Page 1987) were used to initially select highly resistant colonies derived from importations from Russia (Rinderer et al. 2005) and highly susceptible colonies from the United States. Three colonies from three different queen lines founded the resistant Russian population. The susceptible population was derived from three colonies selected from two U.S. commercial sources. In either spring or autumn 2003–2006, surviving colonies from

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both parental populations were screened to select the most divergent sources from which to produce new generations. Matings to produce parental and F₁ colonies were made with instrumental insemination.

Seven groups of test colonies (a group of F₁ colonies with additional colonies representing the two parental groups) were tested using bioassays of young workers (Gary and Page 1987). Four of the seven groups, with F₁ colonies resulting from single drone inseminations, were each tested in a separate single bioassay. The three other groups, with F₁ colonies derived from multiple drone inseminations, were tested in two bioassays each. On average, each bioassay tested 3.5 (range, 2–4) resistant, 4.7 (2–5) F₁, and 3.3 (2–6) susceptible colonies. In total, 25 resistant, 23 susceptible and 27 F₁ colonies were tested.

In each bioassay, young (<12-h-old), uninfested adult bees were obtained as they emerged from individually caged brood combs held in incubators (dark, 35°C, 50–80% RH). Each of 20–50 bees per colony was coded to colony source by marking with a 1-mm dot of gloss enamel paint on the posterior abdominal tergites. Marked bees were placed into the brood nests of inoculation colonies that had 30–90% of resident bees infested with tracheal mites. Marked bees were retrieved from inoculation colonies after 5 to 7 days and then stored frozen until the prothoracic tracheal trunks were dissected and newly infesting adult female mites between the spiracle and first tracheal bifurcation were counted. Average mite abundance (female mites per worker) was calculated for each colony in each bioassay and used as the variable for analyses. This variable was analyzed using a randomized block design with bee type (resistant, F₁ or susceptible) as a fixed effect, and group as a random effect (SAS Institute 2000). Additionally, each of the seven groups was analyzed separately to produce mean separation between the three bee types.

In July 2005, a group of 27 selected Russian resistant, 17 F₁, and 28 selected susceptible multiply-inseminated queens were randomly assigned to colony divisions and established in two apiaries near Baton Rouge, LA. Initial colony infestation was highly variable but evenly distributed between the three groups. Tracheal mite infestations were sampled and the queen status was checked bimonthly until May 2006. Forty bees from each colony were dissected to calculate mite prevalence (percentage of bees infested). Mite growth in colonies was initially slow and queen losses were high. Initial and final mite prevalences from 17 colonies with original queens in May 2006 are presented.

Results and Discussion

The average mite abundance of bees from F₁ colonies in bioassays was intermediate (1.04 ± 0.13 mites per bee; least squares mean ± SE) between that of resistant Russian (0.74 ± 0.13) and selected susceptible (1.57 ± 0.13) parental colonies. The rankings in means from each bee type within each test group were generally consistent (Fig. 1), leading to a low inter-

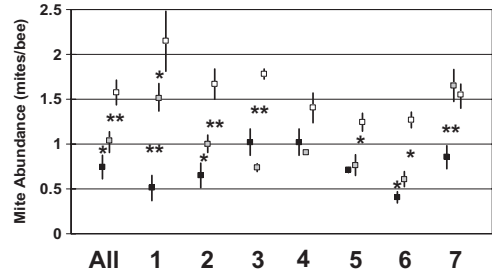


Fig. 1. Mean abundance (±SE) of female tracheal mites in workers from seven groups of colonies of three bee types (selected resistant Russian, solid squares; selected susceptible, open squares; and F₁, gray squares). The results of all colonies considered simultaneously with group as a random block are indicated (All), and an overall significant difference between the F₁ colonies and parental colonies is indicated. Results within each group (1–7) analyzed individually are also indicated. F₁ colonies in groups 1–4 had queens with single drone inseminations, and the other three groups had queens inseminated with multiple drones. Significantly different means between F₁ colonies and corresponding parental colonies in each group are shown (* indicates 0.01 < P < 0.05 and ** indicates P < 0.01 for one-tailed tests of significance).

action between bee type and group (Table 1). The use of single or multiple inseminations to produce F₁ colonies did not appreciably influence the patterns of mean separation (Fig. 1).

Similar infestation trends were evident in the field colonies monitored for 10 mo (Fig. 2). Final tracheal mite infestations in F₁ colonies were intermediate between those of the parental populations. Mites were not detected in any of the three selected Russian colonies with original queens. Mite prevalences ranged from 0% to 53% in bees from four F₁ colonies and from 0 to 90% in 10 susceptible colonies.

These results are in general agreement with other findings assessing the level of resistance of hybrids using different resistant and susceptible populations (Calvert 1957, Lin et al. 1996, Danka and Villa 2000). Tracheal mite resistance is clearly heritable and typically improves the level of resistance in a first generation cross with a susceptible source. The degree to which F₁ colonies resemble resistant parental populations is useful to interpret the genetics of resistance. Field infestations of hybrid colonies best fit a pattern derived from a single dominant gene for resistance (Calvert 1957, Rinderer et al. 1993). Bioassays with Buckfast bees as the resistant parent suggest partial or

Table 1. Analysis of variance structure and results for mite abundance (female mites per worker) in seven groups of propagated colonies representing three bee types (selected resistant Russian parental, selected susceptible parental and F₁ colonies)

Source	Fixed effects			Random effects	
	df	F	P > F	Source	Variance
Bee type	2, 12	12.74	0.0011	Group	0.0272
				Bee type × group	0.0713
				Residual	2.2872

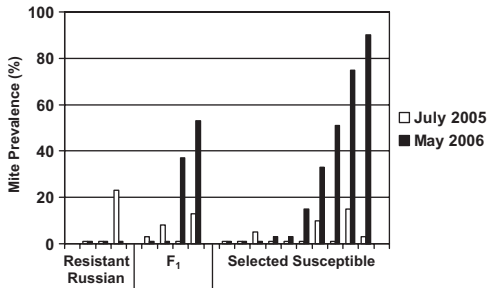


Fig. 2. Changes in tracheal mite prevalences in selected resistant Russian ($n = 3$), F_1 hybrid ($n = 4$) and selected susceptible ($n = 10$) colonies established in July 2005 from colony divisions and maintained in two apiaries in Baton Rouge, LA. In total, 72 colonies were established initially, and only 17 had original queens in May 2006. All colonies were headed by queens inseminated with semen from multiple drones.

incomplete dominance (Lin et al. 1996, Danka and Villa 2000). Although the different findings may reflect the particular genetics of the populations, they also may be influenced by analysis and interpretation. Specifically, when the seven groups in this test are analyzed individually, mean separations differ (Fig. 1). In three groups, F_1 colonies are intermediate and significantly different from either parent. These results, together with the general trend that F_1 colonies are numerically more susceptible than their resistant parent suggest that other loci in addition to the hypothesized major locus affect the strength of the expression of resistance. In one of the seven groups, F_1 colonies are statistically similar to the resistant Russian population, suggesting a major dominant effect favoring resistance. In one group, F_1 colonies are similar to the susceptible colonies. This result, together with the apparent dominance favoring resistance, suggests a single locus and that the resistant parent was heterozygous. In one group, there were no significant differences between the types, possibly because the parental colonies were not very divergent.

The results from bioassays and the field test support the concept that tracheal mite resistance in Russian bees is determined by complete dominance at a single locus and the addition of enhanced resistance from additive genes. Russian bees use autogrooming to remove mites significantly better than their resistant parent (Villa 2006). This behavior also was found to largely explain resistance in Buckfast bees (Danka and Villa 1998) and in selected U.S. colonies (Pettis and Pankiw 1998). Autogrooming is a complex behavior likely regulated with some genes having larger and some having lesser influence on the behavioral expression.

Selected resistant Russian bees are a valuable tool to deal with the persistent problems with tracheal mites experienced by beekeepers, particularly in colder climates that promote the rapid development of infestations (De Guzman et al. 2002, 2006). Using selected Russian queens from commercial queen production enterprises promoting production of similarly resistant drones will guarantee lack of problems from tra-

cheal mites. Where this is not possible or during transitions in the genetics of commercial populations, first generation hybrid colonies can mitigate problems.

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