COMPARATIVE SUSCEPTIBILITY OF AFRICANIZED HONEY BEES[1] FROM SOUTH TEXAS TO INFESTATION BY *ACARAPIS WOODI*[2]

Robert G. Danka and José D. Villa

Honey Bee Breeding, Genetics and Physiology Laboratory
Agricultural Research Service, U.S. Department of Agriculture
1157 Ben Hur Road, Baton Rouge, LA 70820

ABSTRACT

The proportion of Africanized honey bees (*Apis mellifera scutellata* Lepeletier hybridized with European subspecies) that became infested with honey bee tracheal mites (*Acarapis woodi* [Rennie]), the number of female mites per infested bee, and the number of immature mites produced per each test bee were more similar to those of resistant European bees than susceptible European bees when groups of young bees were exposed to mites in infested colonies. Mean mite reproduction in the three bee types did not differ. Thus the mite resistance of Africanized bees was founded on a disruption of the migratory phase but not the reproductive phase of the mite life cycle, as has been found previously when resistant bees were compared with susceptible bees. Although Africanized bees from the southern United States somewhat resist infestation by *A. woodi*, this protection does not exceed that of commercially available European stocks.

INTRODUCTION

Africanized honey bees (*Apis mellifera scutellata* Lepeletier hybridized with European subspecies) now are established in the United States in southern portions of Texas, New Mexico, Arizona and California. The range of Africanized bees continues to expand, especially in the far western areas, and these bees are expected to pose apicultural and public health problems for at least the near future.

Acarapis woodi (Rennie), the honey bee tracheal mite, is an endoparasite that causes weakening and mortality of severely infested bee colonies (Eischen et al. 1989, Otis and Scott-Dupree 1992). Since infestation first was detected in the United States in 1984, it has become apparent that problems in North America are most acute in northern areas where winters are protracted (Otis 1990). Beginning in 1987 (Gary and Page 1987), several types of European-stock bees comparatively resistant to tracheal mite infestation have been identified in the United States and Canada (e.g., Clark et al. 1990, Szabo et al. 1991, Danka et al. 1995, Lin et al. 1996).

The effects of tracheal mites on Africanized bees are largely unknown. Africanized bees infested with the mite have been reported from Brazil (Lenhart et al. 1974) and Costa Rica (Otis et al. 1988). This bee type thus is not immune to parasitism, but mite associated problems seem to be uncommon in the Neotropics. We assessed the susceptibility of southern U.S. Africanized bees to infestation by tracheal mites by directly comparing several features of infestation in

[1]Hymenoptera: Apidae
[2]Acari: Tarsonemidae

Africanized bees with those in two types of European bees: one stock known to be resistant to mite infestation, and one known to be susceptible. Adult female *A. woodi* leave the tracheae in which they developed and migrate into the tracheae of young adult bees to reproduce; female progeny of these foundress mites reach adulthood beginning about 11-14 days after host bees are infested (Bailey and Ball 1991, Pettis and Wilson 1996). Based on this biology, we measured several parameters to evaluate potential differences in infestation between bee types during the migratory and reproductive phases of the life cycle of the parasite.

MATERIALS AND METHODS

The relative susceptibilities of the three bee types was assayed during February-March 1995 by exposing uninfested young adult worker bees from test colonies to tracheal mites in infested inoculation colonies, following procedures similar to those of Gary and Page (1987). Africanized colonies were derived from swarms captured in the Rio Grande Valley of Texas and maintained in two isolated apiaries near Rio Grande City, Texas. Ten test colonies were selected from this pool after morphometric multivariate analyses (Rinderer et al. 1993) indicated a high probability of Africanization [P(A)]: all test colonies had P(A) > 0.95, and five had P(A) > 0.99. Resistant European colonies were of a stock (Buckfast) imported into the United States from the United Kingdom in 1990. Susceptible European colonies were of a stock selected in Louisiana beginning in 1990. These two stocks have been maintained and homogenized by closed breeding. They have shown differential susceptibilities to tracheal mite infestation in field tests (Danka et al. 1995) and in short term evaluations using the bioassay employed here (Danka and Villa 1996). Four colonies of each of the two European bee types were used.

Combs with sealed worker brood were transported either in fiberglass windowscreen bags (Africanized bees from Rio Grande City) or inside holding colonies (resistant and susceptible bees from Baton Rouge, LA) to the USDA-ARS Honey Bee Research Unit at Weslaco, Texas. All adult bees were removed and the combs were held, grouped by colony source, in screen bags inside incubators at 35°C and 50-80% RH.

Newly emerged (0-24 h old) bees were identified as to colony source with coded single or double enamel paint marks (about 1 mm diameter) on abdominal tergites IV or V. Fifty-bee cohorts from each test colony were placed into the broodnests of four inoculation colonies having 43, 48, 68 and 83% of adult bees infested with *A. woodi*. After 10-11 days, marked bees were retrieved into flasks which were kept on crushed ice and filled with CO_2 to immobilize bees and mites; samples later were stored frozen. The numbers of foundress mites and immature mites (progeny) in each trachea of each bee were determined by excising the prothoracic tracheae (spiracle to first bifurcation) from thawed bees and examining them at 30-60X with a dissecting microscope. Male mites occasionally mature in bees as young as 11 days old (Pettis and Wilson 1996); the few males observed thus were classified as progeny. Female mites rarely mature this soon, but if they occurred in our samples this would have led to slight overestimation of foundress populations and underestimation of reproductive rates. Of 72 samples from individual test colonies, 35 were based on 40 bees, 18 had 30-39 bees, 17 had 20-29 bees, and two had 14-17 bees.

Four mite infestation parameters were compared among the three bee types by analysis of variance. Parasitological terminology follows that of Margolis et al. (1982). Prevalence is the percentage of infested bees within a cohort of test bees. Foundress intensity is the number of foundress mites parasitizing individual infested bees Mean fecundity is the average number of offspring produced per foundress mite in each infested bee. Progeny abundance is the number of first generation mites per bee for all bees in a cohort; this measure indicates the immediate future mite threat to a colony. The experiment was analyzed as a split plot design with bee type tested as the whole plot (using colony within bee type as the error term) and

inoculation colony as the subplot (using the interaction of inoculation colony × colony within bee type as the error term). Means of bee type responses within inoculation colonies were separated using least significant differences calculated with weighted error terms that used both whole plot and subplot errors (Cochran and Cox 1957).

RESULTS AND DISCUSSION

The prevalence, foundress intensity and progeny abundance of *A. woodi*, but not the mean fecundity of the parasite, differed among bee types (Table 1). In three of four inoculation colonies, resistant bees and Africanized bees generally had significantly lower tracheal mite prevalences, foundress intensities and progeny abundances than susceptible bees (Fig. 1). These trends were qualitatively similar but not statistically significant in the fourth inoculation colony. When averaged across all inoculation colonies, Africanized bees had 6% proportionally greater prevalence than resistant bees and 22% proportionally lesser prevalence than susceptible bees. Relative differences in foundress intensity among the bee types closely followed differences in prevalence among bee types. Overall foundress intensity in Africanized bees (1.7 ± 0.1 [$\bar{x} \pm$ SEM]) was intermediate between that in resistant bees (1.5 ± 0.1) and that in susceptible bees (2.0 ± 0.1). Progeny abundance in Africanized bees on average was 16% more than in resistant bees and 40% less than in susceptible bees.

The different inoculation colonies used in the four trials influenced the response for each infestation parameter (Table 1). Overall, there was a weak trend for infestations to be greater when prevalences in inoculation colonies were greater. However, infestations in test bees also were affected by other, unmeasured factors within inoculation colonies (e.g., perhaps the age demographics and absolute numbers of resident bees). Statistical interactions of the effects of bee type and inoculation colony occurred for the parameters mean fecundity and progeny abundance. Responses for mean fecundity were variable and difficult to interpret. For progeny abundance, the interaction apparently arose because of disproportionately increasing differences in means at greater inoculation colony prevalences.

Africanized honey bees from south Texas were comparatively resistant to infestation by the honey bee parasite *A. woodi*. These bees were less susceptible to infestation than were bees of a susceptible U.S. stock, but were somewhat more susceptible than bees of a stock known to be resistant from previous studies (Danka et al. 1995, Lin et al. 1996). The measures of progeny

TABLE 1. Results of Analysis of Variance of Effects Influencing Four Parameters of Tracheal Mite Infestation When Africanized, Resistant and Susceptible Honey Bees Were Bioassayed.

Variance source	Prevalence *F*; df; *P*	Foundress intensity *F*; df; *P*	Mean fecundity *F*; df; *P*	Progeny abundance *F*; df; *P*
bee type	6.51; 2, 15; 0.009	7.72; 2, 15; 0.005	2.67; 2, 15; 0.102	10.67; 2, 15; 0.001
inoc. col.	39.30; 3, 45; <0.001	9.39; 3, 45; <0.001	13.13; 3, 45; <0.001	51.51; 3, 45; <0.001
bee type × inoc. col.	2.00; 6, 45; 0.086	0.84; 6, 45; 0.542	2.52; 6, 45; 0.034	4.43; 6, 45; 0.001

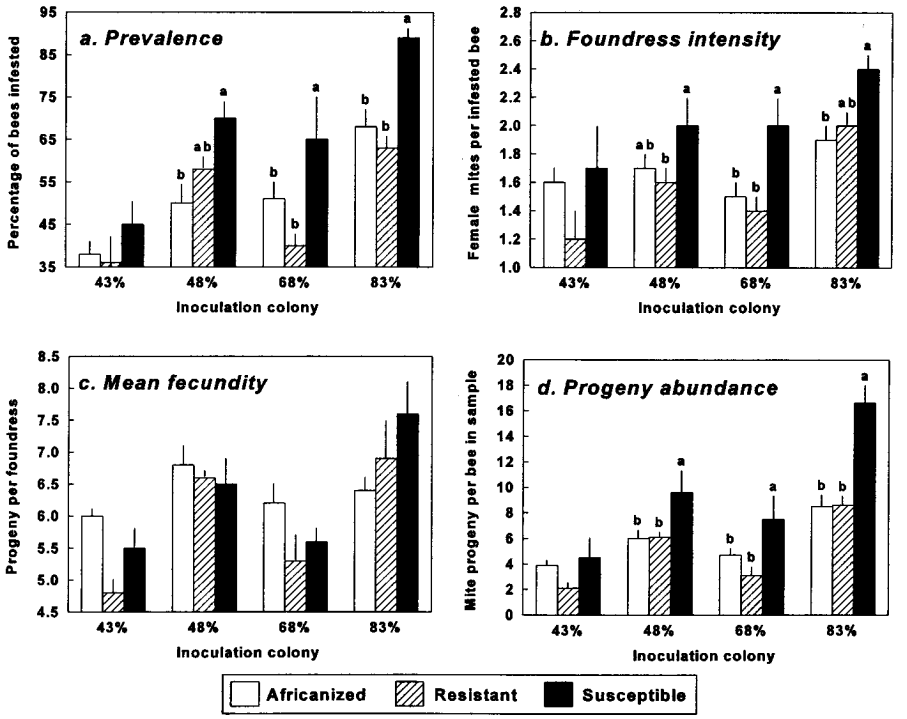

FIG. 1. Means of responses for four parameters of tracheal mite infestation in Africanized, resistant and susceptible honey bees. Error lines at tops of bars indicate one SEM. For each parameter, means within each inoculation colony that are unlettered or share the same letter do not differ at $P > 0.05$.

abundance suggested that comparable initial infestation pressure would be about one-sixth greater of a threat to Africanized colonies than to resistant colonies. However, our study did not assess the proportions of mite progeny that mature and reproduce. The longevity of adult Africanized bees is less than that of European bees in the tropics (Winston 1987) (comparative measurements have not been made in temperate regions). As pointed out by Roubik and Reyes (1984), this factor could further diminish the virulence of infestations in Africanized colonies because mites would have less time to mature.

The general means of resistance of Africanized bees paralleled that found previously for resistant European bees (Danka and Villa 1996). Relative to susceptible bees, tracheal mite migration and establishment (measured as prevalence and foundress intensity, respectively) in resistant bees was suppressed, but reproduction by foundress mites was not. The specific mechanisms regulating this differential mite transfer in bee stocks have yet to be determined.

A. m. scutellata had been under no selection pressure from tracheal mites prior to being exported to Brazil. Indeed, the mites were found only recently in South Africa (Matheson 1996), one of the two African countries of origin of Africanized bees, and still are unknown from Tanzania. Africanized bees, however, apparently have been under some selection pressure from the parasite during much of their existence in the Americas. Tracheal mites were found in Argentina in 1948 (Dyce 1955), Brazil in 1970 (Nascimento 1970), Colombia in 1979

(Menapace and Wilson 1980) and Mexico in 1980 (Wilson and Nunamaker 1982), and were reported from Venezuela in 1982 (Delfinado-Baker and Baker 1982). Most countries of Central America (Belize, Costa Rica, El Salvador, Honduras and Nicaragua) may have become infested with tracheal mites as Africanized bees expanded northward, as these countries were reported to be free of tracheal mites in 1982 (Nixon 1982) but infested in 1993 (Matheson 1993). Otis et al. (1988) suggested that Africanized bees probably vectored the mites into Costa Rica. Thus, Africanized bees may have had sufficient exposure to the mites to result in some resistance being expressed in the bee population expanding northward. However, observations that the parasite is much less of a problem in the relatively low elevation, warmer areas where Africanization is greatest both in the Neotropics (in Brazil for example, Shimanuki et al. 1990) and in the southern United States suggest that selection pressure may have been weak. A second explanation for resistance is that African bees may have been preadapted fortuitously against tracheal mite parasitism by factors such as enhanced grooming ability or cuticular chemistry that renders them less attractive to the mite. A third prospect is that recent hybridization with resistant European bees conferred resistance in the Africanized population we studied. Comparisons of Africanized and European bees existing at the margin of an Africanized zone would address this hypothesis; we did not test local European bees from south Texas.

The practical consequences of Africanized bees being somewhat resistant to tracheal mites are difficult to estimate, but it appears that this parasite will not significantly suppress the feral Africanized bee population of the southern United States. If resistance in Africanized bees is founded on mechanisms currently not widely found in U.S. bees, then the influx of Africanized germplasm may enhance survivorship of feral colonies threatened by tracheal mites. From an apicultural standpoint, the degree of resistance does not exceed that found in European stocks commercially available in North America. Given the notable management difficulties with Africanized bees, these bees should not be considered for use to counter tracheal mites.

ACKNOWLEDGMENT

We thank James Baxter, Arturo Cavazos, Rey Garza (USDA-ARS, Weslaco, TX), Gary Delatte, Lilia de Guzman and James Pursiful (USDA-ARS, Baton Rouge, LA) for assistance in field activities, Chuy Maldonado (USDA-ARS, Weslaco, TX) for morphometric analyses of Africanized colonies, and Anita Collins (USDA-ARS, Weslaco, TX) for use of colonies and facilities. This research was completed in cooperation with the Louisiana Agricultural Experiment Station.

LITERATURE CITED

Bailey, L., and B.V. Ball. 1991. Honey bee pathology, 2nd ed. Academic Press, London.

Clark, K.J., E. Huxter, N.J. Gates, and T.I. Szabo. 1990. Screening breeder honey bee stock for resistance to tracheal mites. Am. Bee J. 130: 800.

Cochran, W.G., and G.M. Cox. 1957. Experimental Designs., 2nd ed. Wiley and Sons, New York.

Danka, R.G., J.D. Villa, T.E. Rinderer, and G.T. Delatte. 1995. Field test of resistance to *Acarapis woodi* (Acari: Tarsonemidae) and of colony production by four stocks of honey bees (Hymenoptera: Apidae). J. Econ. Entomol. 88: 584-591.

Danka, R.G., and J.D. Villa. 1996. Influence of resistant honey bee hosts on the life history of the parasite *Acarapis woodi*. Exp. Appl. Acarol. 20: 313-322.

Delfinado-Baker, M., and E.W. Baker. 1982. Notes on the honey bee mites of the genus *Acarapis* Hirst (Acari: Tarsonemidae). Internat. J. Acarol. 8: 211-226.

Dyce, E.J. 1955. Beekeeping in Peru. Part 2. Am. Bee J. 95: 322-323.

Eischen, F.A., D. Cardoso-Tamez, W.T. Wilson and A. Dietz. 1989. Honey production of honey bee colonies infested with Acarapis woodi (Rennie). Apidologie 20: 1-8.

Gary, N.E., and R.E. Page, Jr. 1987. Phenotypic variation in susceptibility of honey bees, *Apis mellifera*, to infestation by tracheal mites, *Acarapis woodi*. Exp. Appl. Acarol. 3: 291-305.

Lenhart, T., A.S. Michael, and M.D. Levin. 1974. Disease survey of South American Africanized bees. Am. Bee J. 114: 338.

Lin, H., G.W. Otis, M.E. Nasr, and C.D. Scott-Dupree. 1996. Comparative resistance in Buckfast and Canadian stocks of honey bees (*Apis mellifera* L.) to infestation by honey bee tracheal mites (*Acarapis woodi* (Rennie)). Exp. Appl. Acarol. 20: 87-101.

Margolis, L., G.W. Esch, J.C. Holmes, A.M. Curris, and G.A. Schad. 1982. The use of ecological terms in parasitology (Report of an ad hoc committee of the American Society of Parasitologists). J. Parasitol. 68: 131-133.

Matheson, A. 1993. World bee health report. Bee World 74: 176-212.

Matheson, A. 1996. World bee health update 1996. Bee World 77: 45-51.

Menapace, D.M., and W.T. Wilson. 1980. *Acarapis woodi* mites found in honey bees from Colombia, South America. Am. Bee J. 120: 761762, 765.

Nascimento, C.B. 1970. Pesquisa de endo e ecto-parasitos de *Apis mellifera* L, pp. 199-207. *In* 1° Congresso Brasileiro de Apicultura. Florianópolis, SC, Brazil [cited in Apic. Abstr., 1974, 25: 149].

Nixon. M. 1982. Preliminary world maps of honeybee diseases and parasites. Bee World 63: 23-42.

Otis, G.W. 1990. Results of a survey on the economic impact of tracheal mites. Am. Bee J. 130: 28-31.

Otis, G.W., J.B. Bath, and W. Ramirez B. 1988. Have Africanized bees brought honey-bee tracheal mites to Costa Rica?, pp. 541-545. *In* G.R. Needham, R.E. Page, Jr., M. Delfinado-Baker and C. E. Bowman [eds.], Africanized honey bees and bee mites. Ellis Horwood, Chichester, U.K.

Otis, G.W. and C.D. Scott-Dupree. 1992. Effects of *Acarapis woodi* on overwintered colonies of honey bees (Hymenoptera: Apidae) in New York. J. Econ. Entomol. 85: 40-46.

Pettis, J.S. and W.T. Wilson. 1996. Life history of the honey bee tracheal mite (Acari: Tarsonemidae). Ann. Entomol. Soc. Am. 89: 368-374.

Phelan, P.L., A.W. Smith, and G.R. Needham. 1991. Mediation of host selection by cuticular hydrocarbons in the honeybee tracheal mite *Acarapis woodi* (Rennie). J. Chem. Ecol. 17: 463-473.

Rinderer, T.E., S.M. Buco, W.L. Rubink, H.V. Daly, J.A. Stelzer, R.M. Riggio, and F.C. Baptista. 1993. Morphometric identification of Africanized and European honey bees using large reference populations. Apidologie 24: 569-585.

Roubik, D.W. and F. Reyes. 1984. African honey bees have not brought acarine mite infestations to Panama. Am. Bee J. 124: 665-667.

Shimanuki, H., D.A. Knox, and D. De Jong. 1990. Bee diseases, parasites and pests, pp. 283-296. *In* M. Spivak, D.J.C. Fletcher and M.D. Breed [eds.], The "African" honey bee. Westview, Boulder, CO.

Szabo, T.I., L.P. Lefkovitch, and K.J. Clark. 1991. Comparative resistance of honey bees from a closed population to infestation by tracheal mites. Am. Bee J. 131: 643-645.

Wilson, W.T., and R.A. Nunamaker. 1982. The infestation of honey bees in Mexico with *Acarapis woodi*. Am. Bee J. 122: 503-505, 508.

Winston, M.L. 1987. The Biology of the Honey Bee. Harvard University, Cambridge, Mass.