

Evaluating Pure Africanized Honey Bees and Hybrid Crosses for Colony Health and Resistance to Varroa Mites in a Subtropical Climate

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ABSTRACT

Different honey bee, *Apis mellifera* L., breeds were evaluated for overall health and for resistance to the parasitic mite, *Varroa destructor* Oud. in the subtropical Lower Rio Grande Valley (LRGV) in south Texas from June 2005 through October 2006. Breeds examined that have shown genetic resistance to varroa mites were honey bees carrying the Hygienic or SMR/VSH trait (SMR), from Far East Russia (RUS), and the local feral africanized honey bee (AHB). In addition, RUS and SMR crossed with AHB to produce hybrids were also examined. All varroa mite resistant breeds were evaluated against the varroa mite susceptible European (Italian) honey bee breed. To estimate the colony health, the number of adults and the amount of honey produced were recorded monthly. In addition, varroa mite populations in a hive were estimated using a mite-fall count onto a stick board. Typically, the AHB, SMR, and SMR x AHB hybrid breeds produced more adults and honey compared to the Italian breed. Surprisingly, the pure RUS breed had high levels of varroa mites with the fewest adults and low honey production, suggesting that this breed will not perform well in the LRGV. In contrast, while varroa mite populations were somewhat lower than the pure RUS breed, the RUS x AHB hybrid breed clearly had the most adults and eventually produced the most honey compared to all other breeds, including the pure AFB breed. In addition, the SMR x AHB hybrid had the lowest varroa mite populations compared to all breeds, including the pure SMR breed. The fact that these two hybrid breeds performed better than the corresponding pure breeds suggests that heterosis (i.e., hybrid vigor) was observed. Both AHB hybrid breeds appeared to be healthy and contain the fewest varroa mites, but were still as aggressive as the pure AHB breed and difficult to manage. We also examined two types of bottom boards for the affects on varroa mite populations. No significant differences in varroa populations were observed between solid and screen bottom boards for all breeds examined suggesting that this is not a viable method for reducing varroa mite populations in managed hives located in the subtropical LRGV.

Additional Index Words: *Apis mellifera*, Heterosis, parasites

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Worldwide Integrated Pest Management (IPM) strategies are clearly needed to improve the overall health and sustainability of managed honey bee, *Apis mellifera* L., colonies. To date, IPM studies conducted in several different regions of North America have only reported mixed results, including success of these integrated strategies in reducing the devastating parasitic mite, *Varroa destructor* Oud. (Rice et al., 2004; Sammataro et al., 2004; Delaplane et al., 2005).

Varroa mites have developed resistance to both fluralinate and coumaphos which were both widely used for controlling varroa mites throughout the United States (Elzen, et al., 1998; Elzen and Westervelt, 2002); however, commercial beekeepers have raised questions about the practicality of adopting certain miticide-resistance strategies. Furthermore, a beehive modification using screened bottom boards has also been demonstrated to reduce

varroa mite populations (Pettis and Shimanuki, 1999; Harbo and Harris, 2004), but with mixed results. Ledezma (2000) and Aumeier (2001) demonstrated that individual bees in Africanized bees (AHB) colonies more readily mechanically removed or groomed varroa mites from their bodies than European bees (Italian), and consequently fall through the screen and out the hive. However, in other studies, significant reductions in varroa populations were not detected when using screened bottom boards (Ellis et al., 2001; Rice et al., 2004).

An important element of an integrated pest management system (IPM) for controlling parasites of honey bees is the use of genetically-resistant breeds. Specifically, several studies have shown genetic resistance to varroa mites including honey bees carrying the Hygienic or SMR/VSH trait (SMR) and bees introduced from Far East Russia (RUS) (Spivak 1996; Harbo and Harris, 1999; Harbo and Harris, 2003; Harris and Rinderer, 2004). AHB have also shown resistance to varroa mites (Ledezma, 2000); however, evaluating AHB bees and potential hybrid crosses to other genetically-resistant breeds has not been explored. Because honey bees are kept throughout the U.S. under varying climactic conditions, it is also desirable to test candidate resistant breeds in different geographic locations such as the subtropical, Lower Rio Grande Valley (LRGV), Texas where feral AHB are almost exclusively found. The purpose of this study was to evaluate genetically-resistant breeds for colony health and resistance to varroa mites, especially with AHB and AHB hybrids in a subtropical climate.

MATERIALS AND METHODS

AHB, Italian, and SMR Study

Standard beekeeping techniques were utilized in all experiments. On April 29, 2005, honey bee colonies (Italian) were established from 2.5 lb. packages at the Welder Wildlife Refuge, located near Corpus Christi, Texas. Colonies (17 per apiary) were established in 3 locations within a one-mile radius of each other. One of four types of queen breeds (Pure Italian and SMR breeds as well as two hybrids: SMR daughter queen open mated to local AHB drones and AHB daughter queen open mated to the same population of local AHB drones) were introduced into each of the packages. Half of the colonies were established in colonies with a screen bottom board and the other with solid bottom boards as according to Pettis and Shimanuki (1999). A 2-gallon top feeder pail of 55% HFCS was fed to all colonies at the time of installation.

Starting in late June 2005 and concluding in May

2006, bee colony strength (i.e. amount of honey and adults) and varroa mite populations were determined monthly, except for June 2006. For each colony, each frame was removed and the number of full-frame equivalents of adult bees and the number filled with honey were counted. Varroa mite populations were estimated by counting the number of varroa mites that fell to and were trapped on a sticky board placed on the bottom board of the hive for three days.

Russian Bee Study

From July through October 2006, two additional bee breeds (RUS and RUS x AHB hybrid) were examined for colony strength and resistance to varroa mites as described above. All colonies (10-16) were developed from packages using methods described above and placed in 5 different locations within a one-mile radius of each other on the study area.

Statistics. In the AHB, Italian, and SMR study, data collection began in June 2005, and all measured variables (i.e. varroa mites, honey and adults) were collected once a month for twelve months, ending in May 2006. The data was then structured to include the variable, time, starting with initial month of data collection and ending with the final month. Analyses of all dependant variables were performed using the GLIMMIX procedure in SAS (version 9.1.3). The counts of the varroa mites are a Poisson distribution, and this probability distribution was specified as such in the model, using the default link function, log. The distributions of adults and honey were Gaussian (normal), and no generalizations were required. The fixed effects in each model were bottom type, breed, and time, and their 2nd order interactions. Time is the repeated measurement, with its subject the colony (or interaction of bottom type*breed*apiary), and first order autoregressive covariance structure. The Kenward-Roger degree of freedom method was used. Estimated means, standard errors, and differences of means were calculated using the LSMEANS option.

In the Russian Bee study, data collection began in July 2006, and all measured variables (i.e. varroa mites, honey and adults) were collected once a month for four months, ending in October 2006. As in the previous study, the data was structured to include the variable, time, starting with initial month of data collection and ending with the final month. Analyses of all dependant variables were performed using the GLIMMIX procedure in SAS (version 9.1.3). The counts of the varroa mites are again a Poisson distribution, and this probability distribution was specified as such in the model, using the default link function, log. The distributions of adults and honey were Gaussian (normal), and no generalizations were required. The fixed effects in each model were bottom

type, breed, and time, and their 2nd order interactions. Time is the repeated measurement, with its subject the colony within each apiary. The first order autoregressive covariance structure was the best fit for the varroa count variable, whereas the unstructured covariance structure fit the ‘adults’ and honey variables. The Kenward-Roger degree of freedom method was used in all variables’ models. Estimated means, standard errors, and differences of means were calculated using the LSMEANS option.

RESULTS AND DISCUSSION

AHB, Italian, and SMR Study

Adults. Over time, as anticipated the number of adult bees significantly increased ($P < 0.05$) for all breeds (Table 1). No significant differences ($P > 0.05$) were observed for type of bottom board. However, significant differences were observed among the breeds and varied across time (Fig. 1). Typically, AHB, SMR x AHB, and SMR breeds had greater numbers of adult bees compared to the Italian breed (Mean \pm SE: AHB, 14.6 ± 0.48 ; SMR x AHB, 14.1 ± 0.43 ; SMR, 12.1 ± 0.47 ; Italian, 10.2 ± 0.31). This became more prominent during the early spring period, where the numbers of adults from the Italian breed did not increase while the other breeds did increase.

Honey. As observed for adults, the amount of honey significantly increased ($P < 0.05$) for all breeds over time (Table 1). The type of bottom board utilized had a significant affect on the amount of honey produced. Colonies that utilized screened bottom boards produced significantly less honey than colonies utilizing solid bottom boards. In addition, although there were no significant differences ($P > 0.05$) in the amount of honey produced across the breeds (Mean \pm SE: SMR x AHB, 6.7 ± 0.36 ; SMR, 5.9 ± 0.34 ; AHB, 5.7 ± 0.37 ; Italian, 4.7 ± 0.22), a significant interaction was observed for time and breed (Fig. 1). As observed for the number of adults, the Italian breed produced less honey during the early spring, while the other breeds had increased honey production.

Varroa. As observed for adults and honey, varroa populations increased ($P < 0.05$) for all breeds over time (Table 1). Although there was a significant time by type of bottom board interaction, there were no significant differences observed for the main effects including the type of bottom board and breed (Mean \pm SE: Italian, 71.8 ± 13.29 ; AHB, 66.0 ± 13.86 ; SMR, 51.5 ± 9.94 ; SMR x AHB, 45.0 ± 8.89).

In a subtropical environment, it appears that these honey bees bred for some resistance to varroa mites are healthier than the susceptible Italian breed. Although it was difficult to distinguish between AHB, SMR, and SMR x AHB breeds for the number of

Table 1. Effect of Bottom, Breed, Time, and Various Interactions on Measured Variables for the AHB, Italian, and SMR Study (Fig. 1.)

| Type III Tests of Fixed Effects | | | | |
|---------------------------------|--------|--------|---------|--------|
| Effect | Num DF | Den DF | F Value | Pr > F |
| Adults | | | | |
| Bottom | 1 | 45.58 | 0.09 | 0.7622 |
| Breed | 3 | 45.57 | 3.67 | 0.0189 |
| Breed*Bottom | 3 | 46 | 0.26 | 0.8548 |
| Time | 11 | 334.8 | 14.75 | <.0001 |
| Time*Bottom | 11 | 334.8 | 0.45 | 0.9346 |
| Time*Breed | 33 | 360.1 | 1.82 | 0.0046 |
| Varroa | | | | |
| Bottom | 1 | 67.65 | 0.11 | 0.7403 |
| Breed | 3 | 67.8 | 1.52 | 0.2166 |
| Breed*Bottom | 3 | 73.94 | 0.83 | 0.4803 |
| Time | 11 | 355.2 | 28.18 | <.0001 |
| Time*Bottom | 11 | 353.5 | 2.43 | 0.0063 |
| Time*Breed | 33 | 357.9 | 1.32 | 0.1162 |
| Honey | | | | |
| Bottom | 1 | 34.61 | 10.94 | 0.0022 |
| Breed | 3 | 34.51 | 2.04 | 0.1264 |
| Breed*Bottom | 3 | 36.25 | 1.00 | 0.4024 |
| Time | 11 | 321.3 | 33.09 | <.0001 |
| Time*Bottom | 11 | 321.2 | 1.41 | 0.1511 |
| Time*Breed | 33 | 350 | 1.74 | 0.0086 |

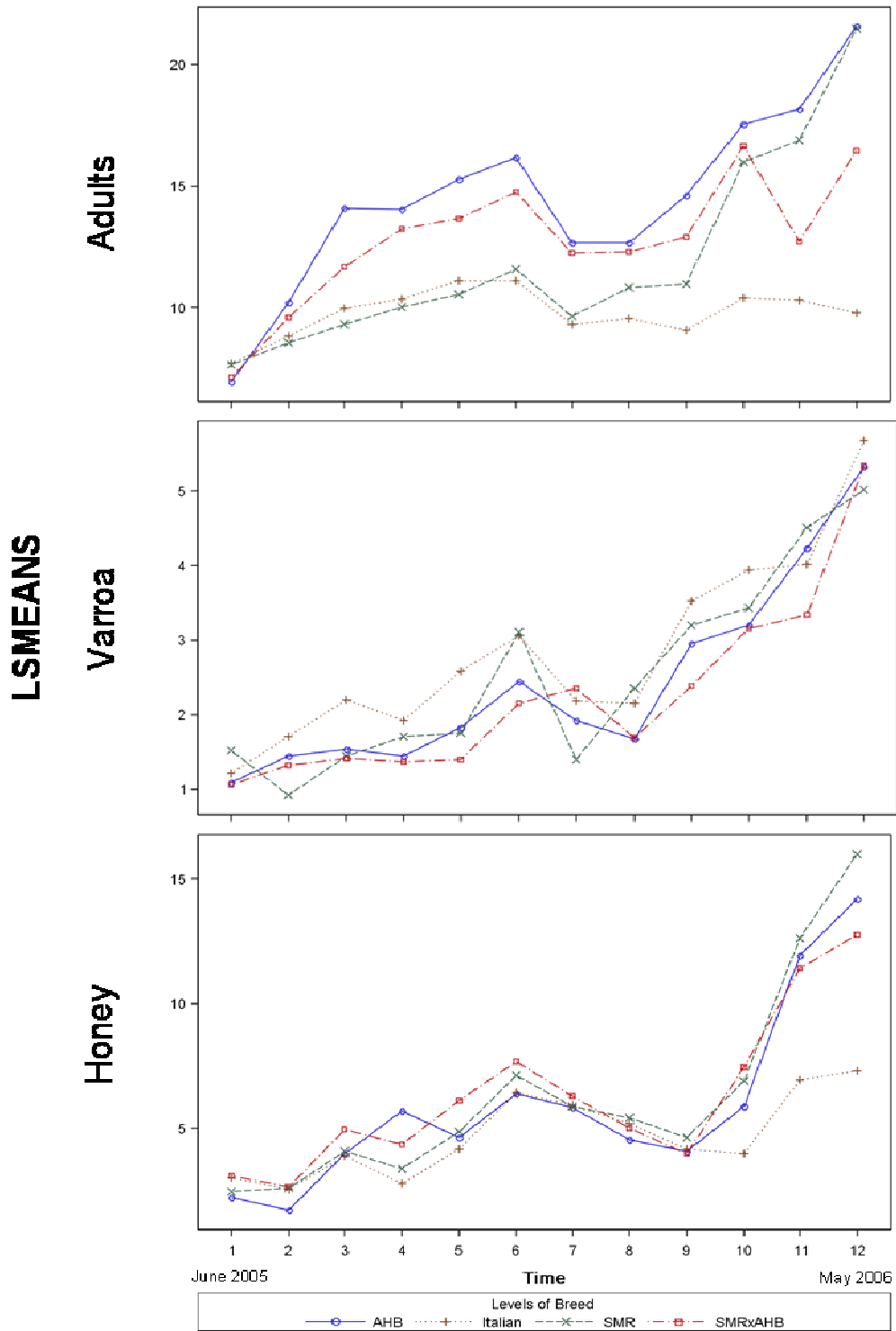


Fig. 1. Population growth of adult honey bees and varroa mites, and honey production from May 2005 to June 2006 for various breeds examined in the AHB, Italian, and SMR Study. AHB = Pure Africanized bee, Italian = Pure Italian bee, SMR = Pure Varroa Sensitive Hygienic bee, and SMR x AHB = Pure Varroa Sensitive Hygienic / Pure Africanized Hybrid bee.

adults and honey produced, these breeds produced more than what was observed for the Italian breed, especially in early spring, when adult populations and honey production typically peaks in the LRGV. Varroa mites were highest for the Italian breed at the beginning of the experiment, further suggesting that this breed was the most susceptible to the parasite. It should be noted that estimating varroa populations across breeds with different mechanisms of varroa mite resistance may be misrepresented using a varroa drop count. For example, varroa mites could have been physically removed by a grooming trait (i.e. SMR breed), and thus these varroa mite populations would appear to be increasing (i.e. dead mites collected on a sticky trap), while the overall health of the colony would also be maintained or even improved (e.g. SMR). Likewise, varroa mite populations could be increasing due to decline of the health of a susceptible breed (i.e. Italian), and thus both SMR and Italian breeds would appear to have similar varroa populations, but with very different colony health.

Russian Bee Study

Adults. As in the previous study, the number of adult bees significantly increased ($P < 0.05$) for all breeds over time (Table 2). However, no significant differences ($P > 0.05$) were observed for type of bottom board. Significant differences were observed in the number of adult bees across the breeds (Mean \pm SE: RUS x AHB, 15.7 ± 0.84 ; AHB, 14.2 ± 0.62 ; SMR x AHB, 13.1 ± 0.77 ; Italian, 12.9 ± 0.65 ; SMR,

12.3 ± 0.67 ; RUS, 10.7 ± 0.43). At each time period, the Russian breed crossed with AHB had higher numbers of adults than any other breed. In contrast, the pure Russian breed had the lowest numbers of adult bees for all examined periods (Fig. 2).

Honey. As in the previous study, the amount of honey significantly increased ($P < 0.05$) for all breeds over time (Table 2). In contrast to the previous study, no significant differences ($P > 0.05$) were observed for type of bottom board. Significant differences were observed in amount of honey produced across the breeds (Mean \pm SE: RUS x AHB, 6.8 ± 0.47 ; SMR x AHB, 5.3 ± 0.49 ; SMR, 5.1 ± 0.40 ; AHB, 4.8 ± 0.34 ; RUS, 4.3 ± 0.25 ; Italian, 4.0 ± 0.39).

Varroa. As observed in the previous study, varroa populations increased ($P < 0.05$) for all breeds over time (Table 1). There were no significant differences observed between the types of bottom board utilized. However, there were significant differences in the number of varroa mites across the breeds (Mean \pm SE: RUS, 29.8 ± 5.58 ; AHB, 24.4 ± 5.29 ; Italian, 21.3 ± 5.72 ; SMR, 17.6 ± 4.25 ; RUS x AHB, 15.2 ± 3.47 ; SMR x AHB, 4.65 ± 1.12). The SMR x AHB breed had the fewest number of varroa mites compared to the other breeds for all examined time periods.

In this study, the effect of breed was significant on all measured variables. Surprisingly, the pure RUS breed had high levels of varroa mites with the fewest adults and low honey production. In contrast, while varroa mite populations were somewhat lower than the pure RUS breed, the RUS x AHB hybrid breed clearly

Table 2. Effect of Bottom, Breed, Time, and Various Interactions on Measured Variables for the Russian Bee Study (Fig. 2.)

| Type III Tests of Fixed Effects | | | | |
|---------------------------------|--------|--------|---------|--------|
| Effect | Num DF | Den DF | F Value | Pr > F |
| Adults | | | | |
| Bottom | 1 | 57.90 | 0.01 | 0.9737 |
| Breed | 5 | 57.82 | 3.34 | 0.0087 |
| Breed*Bottom | 5 | 57.82 | 2.07 | 0.0826 |
| Time | 3 | 51.92 | 19.66 | <.0001 |
| Time*Bottom | 3 | 51.92 | 0.55 | 0.6516 |
| Time*Breed | 15 | 94.92 | 1.50 | 0.1201 |
| Varroa | | | | |
| Bottom | 1 | 72.66 | 1.85 | 0.1780 |
| Breed | 5 | 71.91 | 2.37 | 0.0481 |
| Breed*Bottom | 5 | 71.91 | 1.50 | 0.1997 |
| Time | 3 | 240 | 8.01 | <.0001 |
| Time*Bottom | 3 | 240 | 3.84 | 0.0103 |
| Time*Breed | 15 | 181 | 0.96 | 0.4991 |
| Honey | | | | |
| Bottom | 1 | 61.66 | 0.84 | 0.3618 |
| Breed | 5 | 61.52 | 3.73 | 0.0052 |
| Breed*Bottom | 5 | 61.52 | 1.93 | 0.1024 |
| Time | 3 | 56.29 | 37.44 | <.0001 |
| Time*Bottom | 3 | 56.29 | 1.30 | 0.2824 |
| Time*Breed | 15 | 100.2 | 0.89 | 0.5759 |

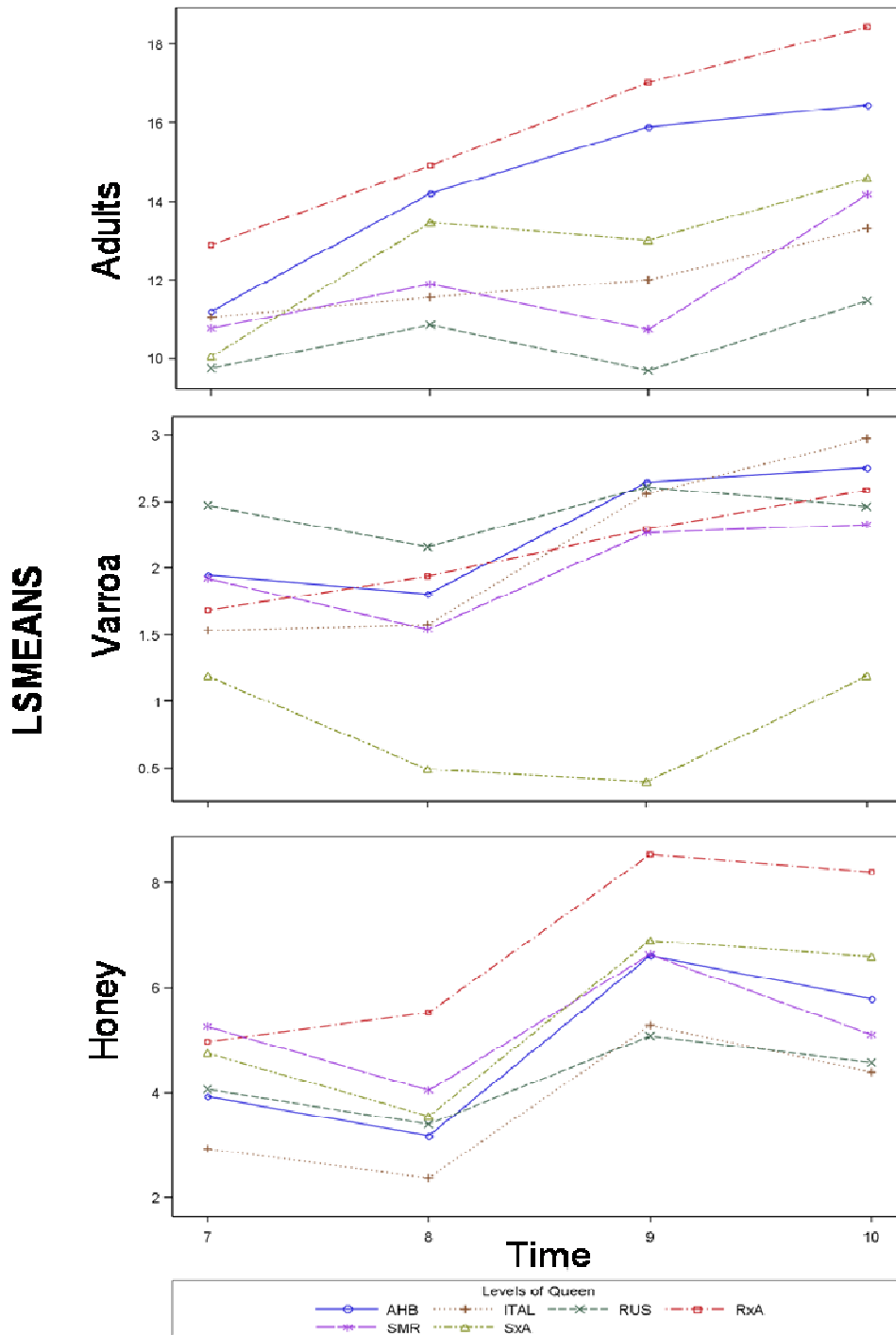


Fig. 2. Population growth of adult honey bees and varroa mites, and honey production from May June 2006 to October 2006 for various breeds examined in the Russian Bee Study. AHB = Pure Africanized bee, Italian = Pure Italian bee, SMR = Pure Varroa Sensitive Hygienic bee, RUS = Pure Russian bee, S x A = Pure Varroa Sensitive Hygienic (SMR) / Pure Africanized Hybrid bee (AHB), and R x A = Pure Russian (RUS) / Pure Africanized Hybrid bee (AHB).

had the most adults and eventually produced the most honey compared to all other breeds, including the pure AHB breed. Unlike the climatically adapted feral AHB, our data suggests that the pure RUS honey bee will not perform well in the subtropical LRGV, perhaps due to climatic differences from the geographic origin of the breed. In addition, the SMR x AHB hybrid had the lowest varroa mite populations compared to all breeds, including the pure SMR breed. The fact that these two hybrid breeds performed better than a corresponding pure breed suggests that heterosis was observed as suggested by Cale et al. (1955) where they defined heterosis or hybrid vigor in the honey bee as the superiority of the hybrid over a parent.

The fact that no significant differences in varroa populations were observed between solid and screen bottom boards for all breeds examined suggests that this is not a viable method for reducing varroa mite populations in managed hives located in the subtropical LRGV. It is important to note that although both AHB hybrid breeds appeared to be very healthy and contain the fewest varroa mites, they were still as aggressive as the pure AHB breed, and were very difficult to manage. However, our data suggests that hybrid AHB breeds can offer a viable option for managing varroa mites, but further selections are needed to reduce their ill-temperament before commercialization.

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