

Novel Pest Management Approach to Manage *Varroa jacobsoni*

Introduction

Because of development of resistance to chemical control by *Varroa jacobsoni* (Elzen 1999, Millani 1999, Vedova et al. 1997), an experiment was designed to test alternative management methods that could be useful in any size of beekeeping operation. In designing this integrated approach, tactics were chosen that were shown to moderate mite levels but were unable to reduce levels singly. The strategies selected for this study included mite-reducing bee stock (queens), screened inserts (screens), and TM07 (active ingredient: thymol; SAFE Inc. Tucson, AZ). Environmental differences between hives were also explored. The queens, screens and environmental factors offer the potential to reduce the population growth rate of mites through multiple mechanisms while the thymol provides the ability to kill adult mite when they have reach critical levels.

Mite reducing bee stock may exhibit hygienic behavior by nurse bees or change in the reproductive rates of mites (Spivak & Downey 1998, Boecking & Spivak 1999, Spivak & Reuter 1998, Spivak 1996, Harris & Harbo 1999). Screened inserts may influence mite population levels by preventing mites from coming in contact with bees (Pettis & Shimanuki 1999) or by altering ambient conditions in the hive. Mites do not retain water well and must imbibe bee hemolymph or take up water vapor to rehydrate (Yoder & Sammataro 1999). The use of thymol to reduce mite levels has been extensively explored in Europe (Imdorf et al. 1999). The work on TM07 is to field test thymol application methods and rates for conditions in the United States.

To know when the mite reducing management strategies are no longer being effective, it is necessary to monitor mite levels in colonies. In the United States differences in the economic threshold for mites have been observed. Delaplane and Hood (1999) estimated for the southeastern United States a threshold of 59-187 mites on overnight sticky boards. However, Caron (1999) found that 30 mites/day on a three-day sticky board may be a better threshold value in the U.S. mid-Atlantic region. Both of these values are substantially higher than the threshold values used in Europe.

The hypothesis that combined strategies would be effective in moderating mite levels was tested in this experiment. Colonies with the lowest mite drop on sticky boards at the end of the experiment indicated that the combination of tactics was successful.

Methods

In May 1999, fifty nucleus colonies were established. The colonies were divided equally between two apiaries approximately five kilometers apart. The Sheltered Site was located in a clearing in a secondary growth forest, which offered the apiary protection from the wind. The Open Site was near the top of a hill in a field subject to constant air movement. The hives at the Open Site were arranged such that half of the hives were positioned with the entrances facing south/southeast and half were facing north. Hives at the Sheltered Site were all facing south. At each apiary, five colonies were randomly assigned to each treatment: control, queen/TM07, screen/TM07, queen/screen, and queen/screen/TM07.

During the first week of June, screen inserts (Brushy Mt. Beekeeping Supply, NC) and mite reducing queen stock (Heikham Queens, CA and Bee Breeding & Genetics Lab, Baton Rouge, LA) were installed. All colonies were adjusted to have similar bee populations in early July.

To monitor mite populations, a newly-designed sticky board (30% of the board is counted; Ostiguy & Sammataro 2000) was used. The collection boards (Great Lakes IPM, MI) were smeared with

petroleum jelly and placed beneath the screen insert under an 8-mesh wire sheet hardware cloth supported with wooden strips to keep the wire off the paper. Pretreatment three-day sticky boards were installed on 7 July and 23 August.

TM07 plastic strips were placed in the thymol treatment groups on 30 August and new three-day sticky boards were inserted in all colonies. At approximately ten-day intervals, post TM07 (thymol) treatment, new three-day sticky boards were installed in all treatment groups. Apistan® strips were inserted into all colonies along with a fresh four-day sticky board on 21 October. At the end of the study (October) the number of frames with bees were counted in each colony to assess colony strength.

Statistical analysis

Multi-factor, block, and repeated measures ANOVA tests were used to analyze location, hive entrance direction and treatment effect on mean mite drop and number of frames with bees (Zar, 1999). The pretreatment natural mite drop obtained on 7 July was used as a covariate in the analysis. Multiple comparison tests were used to evaluate differences among treatment groups (Zar, 1999). Differences between mite-reducing stock were compared by mean mite drop per day and number of frames with bees using a Student's t-test. The statistical package used for data analysis was SPSS (SPSS, 1989-1995).

Results

One month after the nucs were installed, there was no significant difference in mite drop among treatment groups; ANOVA = 1.05; $p=0.38$.

Hive Entrance Direction Effect - North versus South/Southwest

North facing colonies at the Open Site were observed to have larger mean mite drops per day than south/southwest facing colonies (9 September: $p=0.035$ and 21 October: $p=0.027$). There were no north facing colonies at the Sheltered Site.

The number of frames with bees (21 October) was compared by colony entrance direction. Colonies facing north had fewer frames with bees than colonies facing south/southwest ($p=0.005$).

Location Effects

When the natural mean mite drop was compared between treatment groups on 9 September, no significant difference was observed at the Open Site. In contrast, there was a significant difference among treatment groups at the Sheltered Site ($p<0.05$). The queen/screen and queen/screen/TM07 treatment groups were observed to have significantly lower natural mean mite drops than the controls and the queen/TM07 treatments. When the colonies were treated with Apistan on 21 October, a significant difference again was observed between treatment groups at the Sheltered Site but not at the Open Site. The screen/TM07, queen/screen and queen/screen/TM07 treatment groups differed significantly from the control group ($p<0.05$). Additionally, the control and queen/TM07 treated colonies at the Sheltered Site had significantly higher mean mite drops than the corresponding treatments at the Open Site ($p=0.05$ and $p=0.007$, respectively).

The rate of mite population growth was compared by treatment and location. At the Sheltered Site, the queen/screen/TM07, queen/screen and screen/TM07 treatment colonies had significantly slower rates of population growth than the control colonies ($p<0.05$; Figure 1). There was no significant difference in the rate of mite population growth between the treatments at the Open Site. When treatment effects are removed, the overall rate of mite population growth was significantly less at the Open Site than at the Sheltered Site ($p=0.0001$).

The number of frames with bees was compared by hive location. In the Open Site as compared to the Sheltered Site, the number of frames of bees was greater ($p=0.001$). When treatments were compared within each location, no significant differences were observed among treatment groups at the Open Site. In contrast, there was a significant difference among treatment groups at the Sheltered Site ($p<0.05$). The control colonies had fewer frames with bees than the queen/screen and queen/screen/TM07 colonies. When the control colonies at the two sites are compared, the number of frames with bees was significantly less at the Sheltered Site than at the Open Site ($p=0.01$). At the Sheltered Site, control, queen/TM07, and screen/TM07 colonies all have significantly fewer frames with bees than the colonies at the Open Site ($p=0.005$, $p=0.009$, and $p=0.023$, respectively).

A significant inverse correlation was found between mean mite drop after the Apistan treatment and the number of frames of bees ($p=0.0001$). When the effect of the number of bees per colony, location, and hive entrance direction on mite number are removed, a significant treatment effect remains ($p=0.01$).

TM07 Treatment Effects

At the Sheltered Site, the queen/TM07 and queen/screen/TM07 treatment groups were observed to have significantly higher mean mite drops 10 days following the TM07 thymol treatment ($p<0.05$). This increased mite drop continued for the queen/TM07 but not the queen/screen/TM07 treated colonies. The colonies at the Open Site that received the queen/TM07, screen/TM07 or queen/screen/TM07 treatments were observed to have significantly higher mean mite drops 10 days following the TM07 thymol treatment ($p<0.05$). By the end of the study none of the initial differences in mean mite drop between the treatment groups or sites remained.

TM07 treatment did not effect the number of frames with bees in colonies.

Mite-reducing Bee Stock Effect

Two mite reducing bee stocks were used in the study. Throughout the study, the hygienic bee stock had higher mean mite drops but at no time did the differences reach statistical significance. On 15 October and 21 October, the differences in mean mite drop neared significance ($p=0.058$ and $p=0.075$, respectively; Figure 2).

Differences in the number of frames with bees were observed between the two mite-reducing bee stocks. The resistant/tolerant stock was observed to have significantly more frames with bees ($p=0.003$) even though the sample size was very small (resistant/tolerant stock: $n=3$, hygienic stock: $n=23$).

Conclusions

Three strategies were tested in combination with various environmental factors to determine if mite levels in honey bee colonies could be lowered. Two of the prevention tactics tested, mite-reducing bee stock and screen inserts, resulted in significantly lower mite levels. This observation was consistent throughout the summer. We also found that the strategies that were successful at lowering the mean mite drop were inversely correlated with higher numbers of frames with bees. The correlation between lower of mite population size and larger numbers of bees per colony may indicated that the strategies tested reduce the adverse impact of mites on colony health.

Hive location and direction of hive entrance had significant impacts on mite levels. The lowest mite levels were observed in south facing hives colonies at the Open Site. This difference was observed in less than 5 months. The traditional location for honey bee colonies in the Eastern U.S. is a sheltered location with trees nearby. In our study, this location had the highest mite levels. Further work needs

to be done to define and measure the characters of sites to determine the environmental factors that favor colony health and lower mite populations.

The number of frames with bees was correlated with the direction of the hive entrance and hive location. Strongest colonies were located at the Open Site and in colonies with entrances facing south. We also observed that sheltered location hives, even if south facing, do not fair as well as south facing hives in a more open location.

Hive location also influenced our ability to observe treatment effects. The mean mite drop did not differ among treatment groups at the Open Site while significant differences were seen at the Sheltered Site. At the Sheltered Site, the queen/screen and queen/screen/TM07 treatments effectively lowered the mean mite levels. This treatment effect was not observed at the Open Site. The highest mean mite drop observed in a treatment group at the Open Site did not differ from the lowest mean mite drop observed in a treatment group at the Sheltered Site. It seems that the unmeasured environmental factors at the Open Site lower mean mite levels as well, or maybe better, than the treatments. We intend to measure a variety of environmental factors, e.g., temperature, humidity, light intensity, and air speed, to explore possible correlations between environmental factors and mean mite drop.

The effect of location on the number of frames of bees was moderated by treatment. Among the control, queen/TM07 and screen/TM07 treatments, the number of frames of bees was less at the Sheltered than at the Open Site. No location differences were observed among the queen/screen and queen/screen/TM07 groups. The effect of location and treatment on colony strength needs to be explored further.

TM07 caused an immediate increase in mean mite drop but this effect did not result in lower mite drops later in the study. It is possible that the thymol was lost from the hive too rapidly. We intend to measure thymol levels after placement of TM07 strips in the hive to determine if thymol is present and at what concentration.

We found that mite resistant/tolerant bee stock was more successful in reducing mite drop than the hygienic bee stock. Additionally, the mite resistant stock colonies were stronger, as measured by number of frames with bees, than the hygienic bee stock. Because of the very small sample sizes and the severe drought experienced during the study period, we consider these results to be very preliminary. Further work has been planned to explore mite reducing stock differences.

The results from this study of strategies to manage mite levels have been very promising. We intend to continue looking at treatment combinations such as mite-reducing bee stock, screen inserts, and environmental factors that can suppress mite numbers and treatments that can reduce mite numbers when they become elevated.

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Figure 1: Mite population growth rates at two apiary locations by treatment: Mite population growth rates were consistently higher at the Sheltered Site. The mite population growth rate for the queen/screen/TM07, queen/screen/ and screen/TM07 treatments differed significantly from the controls at the Sheltered Site. No significant difference in growth rates by treatment was observed at the Open Site.

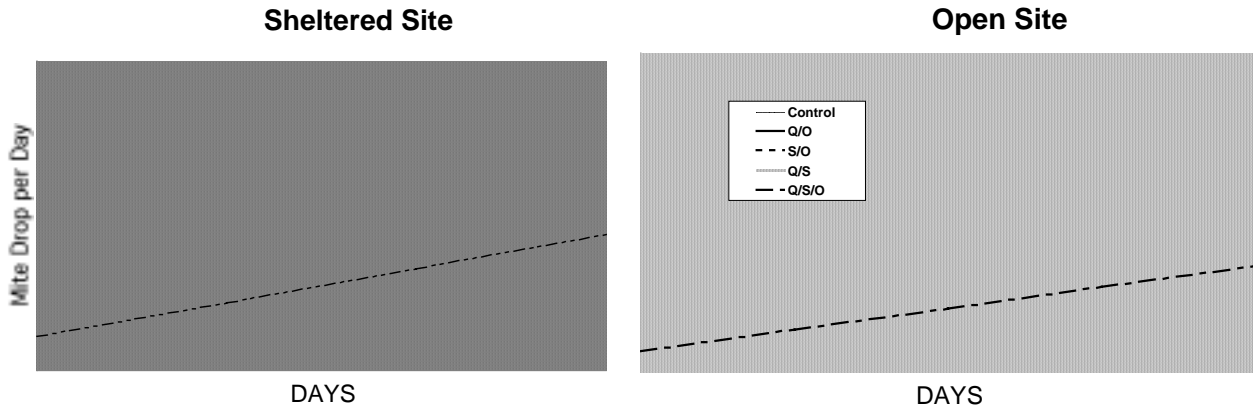


Figure 2: The Effect of Mite Reducing Bee Stock on Mean Mite Drop: Resistant/tolerant bee stock was observed to have consistently lower mean mite drop per day as compared to the hygienic bee stock. On the 125th day of the study (15 October) the mean natural mite drop for the hygienic stock was larger than for the resistant/tolerant stock ($p=0.058$). The difference in the mean mite drop after the application of Apistan on the 131st day (21 October) was not as great ($p=0.075$).

