mites. Bees will open capped brood cells and remove dead or dying brood (Boecking et al 1993, Spivak 1996). Therefore, the drone comb is ready for the queen to lay eggs again after 3-5 days. The main improvement of this method compared to the traditional drone trapping method is that one does not have to open the colony a second time, nor does the drone comb require further handling (being frozen, or uncapped). The "treatment" (applying heat through electricity) can be done multiple times and has a large time window (about 14 days during which the drone cells remain sealed). It is also possible to engineer the Mitezapper to have the heating shut off at 44°C, at that temperature mites die, but drone bees are unharmed (BrØdsgaard and Hansen 1994).

Preliminary results

To prove that this method works, there are several questions that need to be answered. For example, will workers build normal drone wax cells when there is a metal wire embedded? Will the queen lay eggs in such a cell? Will mites invade drone cells built upon wires? How high is the mortality when heat is generated? Will the heat melt the wax when the "electricity" is applied?

To answer these questions I constructed two prototypes. The heating element was regular wires used to strengthen wax foundation. I embedded the wires in the wax foundation with a 12 volt battery charger (6 A), which beekeepers routinely use. Each frame had wires going across 12 times. The two frames were placed into a colony that had not been chemically treated for mites. The workers built normal drone cells on the foundation, cells were sealed normally, and mites invaded these cells. In the first test, with ambient temperature at 27°C, and the average resistance at 1.8 Ohm, it took 5 min to reach 45°C (temperature sensor was put in one pupae), and the mortality was 59%. In a second test, the ambient temperature was 32°C, resistance was 2.0 Ohm, and it took 7.5 min to reach 43°C. I allowed three more minutes of "zapping" after the temperature reached 43°C. This time all 45 mites were dead, a 100% mortality. Mites not subjected to the heat treatment showed a natural mortality of only 9.5% (Fig. 1). (More recent fine-tuning of the method now shows that the entire comb can be heated to the required temperature in about 10 seconds, according to the author.) The device therefore is highly effective in killing mites in a laboratory setting. However, wax near the wires melted down. Clearly, in the final product, the wires have to be embedded inside plastic, preferably plastic that is heat resistant. We are currently conducting field trials to determine whether using this method alone is sufficient to suppress Varroa mite populations so that colonies can survive the winter.

Conclusion

We are currently searching for a manufacturer to produce the Mitezapper. There will be a few engineering problems to be solved, but the principle of using heat generated by foundation wires works. The final product is estimated to be less than $10 a piece and can easily last up to 10 years. The cost would be $1 per colony per year, compared to $8 per colony per year if using chemicals. This would save beekeepers in the U.S. millions of dollars per year with a current estimate of 2.3 million colonies and assuming 50% of the colonies receive chemicals for mite control.

I anticipate that the Mitezapper will be proven to be an effective tool for varroa mite control. The device takes the advantages of the weak point in the mite biological cycle. No chemicals are used, so that honey produced this way is truly "organic".

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The Use of Heat Treatment for Control of the Honey Bee Mite, *Varroa destructor*

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ABSTRACT

The effect of heat treatment on adult worker bees and their phoretic varroa mites was examined at 22°C, 35°C and 40°C. Groups of approximately 50 mite-infested adult bees were subjected to heat treatment and data were collected over 5 days on both mite drop and bee death. Results demonstrate that there is a positive correlation between high temperature (40°C) and increased mite drop. The highest amount of mite drop was seen during the first two days at both 35°C and 40°C, with 40°C producing the greatest amount of mite drop. Bee death at 35°C did not differ from that of our control at 22°C and significant bee death did not occur until after the third day of treatment at 40°C, which was after the period of the greatest mite drop.

INTRODUCTION

Varroa destructor is a serious ectoparasitic mite pest of the European honey bee, *Apis mellifera*. Since its introduction into the United States in 1987, varroa mites have become increasingly problematic to the Western honey bee, which is less tolerant to the mites than the Eastern honey bee, *Apis cerana*, the original host species (Peng et al 1987). Varroa mites, along with the tracheal mite, *Acarapis woodii*, have caused the decline of over 95% of feral honey bees and approximately 30-35% of the managed honey bee colonies in the United States and in many areas world wide.

Currently, there are two general use chemical treatments for varroa mites in the United States, fluvalinate (Apistan®) and formic acid (Apicure™), and one restricted use product, coumaphos (Checkmite®). Fluvalinate is almost 100% effective in killing varroa mites. The chemical is sold as impregnated strips that are placed within the brood chamber of infected hives and acts as a contact pesticide, which is relatively safe for both honey bees and humans if used according to label directions. Apistan® has been available since 1988 and has been quite effective in controlling varroa mites; however, mite populations in the U.S. and abroad have developed resistance to this pesticide (Milani 1999; Elzen et al 1998; Lodesani et al 1995).

Coumaphos was approved in 1998 under an EPA Section 18 Emergency Exemption and is now available in a number of states for use against the small hive beetle, *Aethnia tumida*, as well as for varroa mites. Although shown to be 97-99% effective in killing the mites, coumaphos is an organophosphate and acts as a nerve toxicant to both mites and, in high doses, to humans. Because of its mode of action, the use of coumaphos for mite control is particularly risky due to possible contamination of hive products (wax and honey) intended for human use.

Formic acid, formulated into gel packs and sold under the trade name Apicure™, became available for use in the United States in March 2000 and has been proven to be a safe alternative treatment for varroa mites (Kochansky and Shimanuki 1999; Feldlaufer et al 1997). Formic acid also has the advantage of being effective against tracheal mites. Because formic acid is a naturally occurring substance found in honey, contamination is not a serious concern, but the chemical is only 70% effective in killing varroa mites. The use of formic acid as an alternating treatment to fluvalinate may help slow down the development of varroa resistance.

Even with the approval of formic acid, there are still only a limited number (three) of chemical options available for varroa treatment in the United States. Because varroa mites continue to be a serious threat to the Western honey bee, it is important that effective non-chemical treatments be available as control options. The use of elevated heat treatments has been investigated over the years as a potential control measure for varroa mites. Studies have involved heat treatment of capped bee brood (Marien 1995; Appel and Buchler 1991; Rosenkranz 1987) and of the whole colony or adult bees only (Harbo 2000; McArthur 1990; Hoppe and Ritter 1986; Kommisar 1985). Most of the heat treatment experiments on adult bees involved short temperature spikes at relatively high temperatures. Kommisar (1985) found heat treatment of 47°C for 2-15 minutes to be 97-98% effective in killing mites on adult bees. Hoppe and Ritter (1986) tested temperatures ranging from 42°C - 51 °C, with treatments lasting a maximum of 30 minutes and found that up to 86% of the mites were killed. According to McArthur (1990), treatment of adult bees at 46°C - 48°C for 12-15 minutes would kill 100% of the mites. In an independent but similar study, Harbo (2000) tested temperatures ranging from 25-40°C and found that 100% of the mites dropped from adult bees at 40°C. We examined the effect of five-day heat treatments at 22°C, 35°C and 40°C in mites infesting adult worker bees.

MATERIALS AND METHODS

The experiment was conducted using two 4 ft x 7ft incubation chambers at the NCSU Phytotron facility in which both relative humidity and temperature were controlled (NCSU 1998). Bees were collected from selected hives in the NCSU Apiary. Within each chamber, four boxes of adult bees from a colony heavily infected with varroa mites could be heat-treated for five-day periods. Both mite drop and bee death were recorded for each test box at selected intervals over the course of the five-day treatment.

Standard mailing boxes in which package bees are routinely shipped by the bee industry were modified into test boxes. The wooden boxes had two wire sides, wooden ends, a wooden top and bottom, and a 4-inch diameter hole in the top in which a sugar syrup feeder was placed during transport. Each box was modified to include a piece of 8-gauge hardware cloth that was cut to fit the inside of each box and was attached about 1 1/2- inches from the bottom of the box using craft wire. The hardware cloth served as a platform through which mites could fall, but bees could not

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(Pettis and Shimanuki 1999). Additionally, the periphery of the platform was sealed to the sides of the test box with duct tape to insure that no bees could escape through any gaps on the sides of the cage.

Next, a slit was cut 1/4-inch ftom the bottom across one of the wooden sides of the box using a table saw. A piece of plexiglas cut to fit the bottom of the box was inserted through the slit and could be removed to quantify mite drop. The top of the plexiglas was covered with petroleum jelly and served as a sticky trap to collect mites that fell through the screen platform.

A 3.5 x 3.5-inch square of empty drawn comb was attached on the interior of one of the wooden sides of each box using craft wire. The purpose of the comb was to simulate a hive environment in order to increase overall survival of the bees by reducing bee stress during the test period (Rinderer and Elliot 1977).

Each test box was equipped with a plastic jar feeder with a hole drilled in the lid through which a 6-inch piece of 3/8-inch diameter cotton dental wicking was placed. The feeders were filled with a 1:4 sugar/water solution. Concentrations greater than 1:4 sugar/water resulted in the drying out of the feeding wick. The sugar-delivering wick was inserted through a hole in each test box where the bees could access the wick. The feeders were then duct-taped in an upright position to the outside of each test box.

In order to collect bees for the experiment, several hundred mite-infested adult bees were shaken into an unmodified package box directly from the honey frames of five selected bee colonies. Selected bee colonies had mite/bee levels of from 1:9 to 1:2. The box was then completely covered with brown paper to darken the inside of the box. The bees were allowed to "rest" for 18-24 hours at room temperature (22°C) to allow any mites that may have fallen during transfer to climb back up onto the bees. The rest period also allowed all of the bees to be kept at the same uniform temperature before being tested using temperature treatments. After 24 hours, approximately 50 bees were transferred from the holding box to each of the 8 modified test boxes.

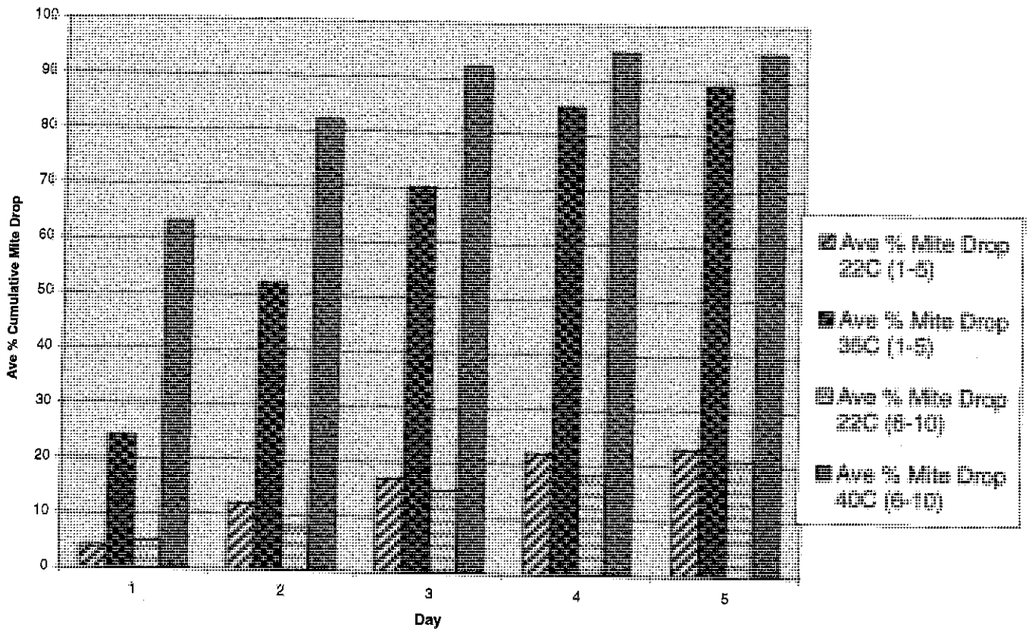
To transfer the bees, transparent vinyl tubing was used as a bridge through which bees could walk undisturbed from the unmodified "parent" box to each of the test boxes. This procedure was used to reduce mite drop/loss that might have occurred if the bees were handled directly. A cardboard square was cut to cover the feeder holes in both the unmodified and modified test boxes. One end of a two-foot piece of 3/4-inch "bridge" tubing was attached to the unmodified package of bees through a hole cut in the cardboard square that was taped over the feeder hole at the top of the box. The free end of tubing was attached to a modified package box through the cardboard in the same manner. A slit was cut a few inches from the opening of the tuhing in the parent box. Using a flashlight to encourage bee movement, the bees were allowed to walk through the tubing undisturbed from the parent box into each modified test box. When the desired number of bees (approximately 50) were transferred, bee travel was blocked with a razor blade inserted into the slit in the tubing. After each test box was filled with the desired number of bees, the cardboard was replaced with a square of plexiglas duct-taped over the hole.

Once all of the test boxes were populated, they were immediately transported to the test chambers in the NCSU Phytotron facility. We utilized two chambers with both temperature and relative humidity controlled for each test run. For experiments 1-5, one chamber was set at 22°C and the other was set at 35°C. For experiments 6-10, one chamber was set at 22°C and the other at 40°C. The temperature settings were alternated with each experiment in order to minimize chamber effects. Twenty-two degrees Celcius is at the lower end of the temperature range at which good bee flight occurs and served as our control. The approximate temperature of a typical brood chamber is 35°C; therefore we knew this temperature would be a safe treatment for bees. We evaluated mite drop at 40°C because it is relatively safe for bees (Appel and Büchler 1991; Engels and Rosenkranz 1992), but may be hot enough to increase mite drop (Harbo 2000). Both chambers were set at 40% RH which is within the of relative humidity range found in a typical *Apis mellifera* colony (Oertel 1949).

The total number of mites present for each group of bees was determined by shaking the bees in 95% ethyl alcohol at the end of each test run. The bees were strained and mites left within the alcohol were counted. Each bee was individually examined for any mites that might not have been successfully removed with the alcohol. The mites collected in the alcohol and individual bee sampling were combined with collected mite drop to determine total mite number for each test box. The daily % mite drops were based on this total mite number.

Four boxes were placed in each chamber. Additional strips of cardboard covered with petroleum jelly were placed around the screen sides of each test box to trap any mites that might fall through the sides of the cage. Experiments 1-5 were conducted from May 1999 to July 1999 and experiments 6-10 were conducted from July 1999 to September 1999. Statistical analysis was performed using a one-sided sign test. Data for both mite drop and bee death were collected over a period of five days for each experiment.

Figure 1. Average % cumulative mite drop at 22°C, 35°C, and 40°C for experiments 1-5 & experiments 6-10.



RESULTS

The data for four individual test boxes per incubation chamber were averaged for each chamber. The procedure was repeated for all ten experiments, producing the following data: % mite drop per day over 5 days for each experiment; % bee death per day over 5 days for each experiment. The data for all of the experiments were combined into two groups before averaging: The first group consists of experiments 1-5 which were tested at 22°C and 35°C and the second group consists of experiments 6-10 which were tested at 22°C and 40°C. Cumulative data are shown in Figures 1 & 2. It should be noted that experiments 1-5 and experiments 6-10 were not conducted simultaneously.

In the first group of experiments, 4% mite drop occurred at 22°C and 24 % mite drop occurred at 35 °C after 24 hours of heat treatment. After 48 hours of treatment 12% and 52 % of the mites dropped at 22°C and 35°C, respectively. After 72 hours of treatment, 17% and 70% of all the mites dropped at 22°C and 35°C. After 96 hours of treatment, there was 22% and 85% mite drop at 22°C and 35°C. After 120 hours of treatment, 23% and 89% of the mites dropped at each respective temperature.

For the second group of experiments at 22°C and 40°C, cumulative average mite drop at 22°C was 5% and at 40°C there was 63% mite drop after the first 24 hours of treatment. After 48 hours, mite drop was 8% at 22°C and 83% at 40°C. After 72 hours of treatment, there was a 14% mite drop at 22°C and 93% mite drop at 40°C. After 96 hours, there was 18% and 95% mite drop at each respective temperature. After 120 hours of treatment, there was 21% mite drop at 22°C and 96% mite drop at 40°C.

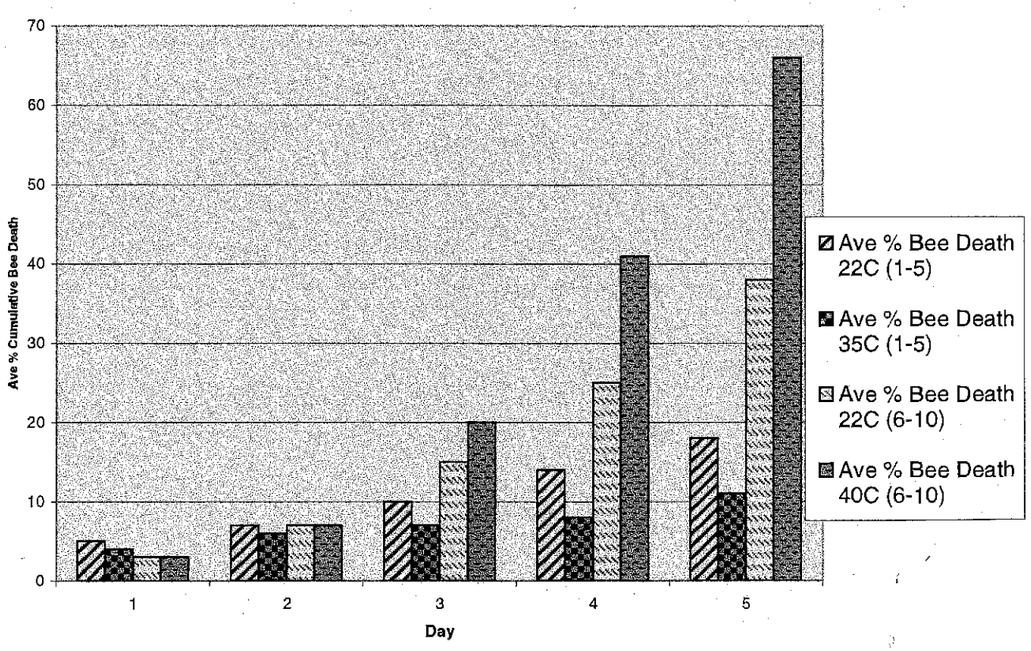
Summary data for bee deaths show that after 24 hours of treatment, only 5% of the bees died at 22°C and 4% at 35°C for the first group of experiments, and 3% at both 22°C and 40°C for the second group of experiments. After 48 hours of treatment, 8% of the bees died at 22°C and 6% at 35°C for experiments 1-5 and 7% of the bees died at both 22°C and 40°C for experiments 6-10. After 72 hours of treatment, 11% and 7% of the bees had died at 22°C and 35°C and 16% and 19% died at 22°C and 40°C. After 96 hours, 15% of bees died at 22°C and 9% at 35°C for the first group of experiments and 26% and 41 % of the bees died at 22°C and 40°C for the second group of experiments. After 5 days of treatment, cumulative bee death percentages were 19% at 22°C, 11 % at 35°C for experiments 1-5 and 36% at 22°C and 60% at 40°C for experiments 6-10.

For mite drop, statistical analysis using a one-sided sign test indicated that all of the experiments had greater significant mite drop at both 35°C and 40°C versus 22°C (P=0.0313). For bee death data, there was no statistical difference among temperature treatments (P>0.05).

DISCUSSION

Mite drop is a natural occurrence caused by factors such as mite behavior (Harbo 2000), bee grooming, hygienic behavior or normal hive activities (Peng et al 1987). Although mite drop occurred at all three test temperatures, we found a significant increase in the proportion of mite drop at both 35°C and 40°C versus that at 22°C. The data indicate that there is a correlation between temperature and mite drop. This reinforces data obtained by Harbo (2000) who obtained comparable, but somewhat higher mite drop levels, using a similar experimental design. Mite drop

Figure 2. Average % cumulative bee death at 22°C, 35°C, and 40°C for experiments 1-5 & experiments 6-10.



increases with rising temperature, with the highest percentage in our experiment occurring at 40°C. Even at 35°C after three days of treatment there was an average of 70% mite drop. Although 35°C is the average temperature in the brood nest, the only bees likely to encounter that temperature on a regular basis are young nurse bees. Our experiment was conducted using older workers which are unlikely to encounter 35°C temperatures inside the hive on a regular basis.

We collected data on bee death at daily intervals for the five days of treatment for 22°C, 35°C and 40°C. Because good bee flight can occur at 22°C, we did not expect to see any significant bee losses due to treatment at this temperature, nor did we expect abnormal bee losses at 35°C, the average temperature of a brood chamber. Our data show only moderate bee loss at both 22 °C and 35 °C as compared to 40°C, and the data at both of these lower temperatures are not significantly different from each other ($P>0.05$). Although there was no statistical significance in bee death among the three temperatures, treatment at 40°C caused a substantially higher % of bee death than the other two temperature treatments after three days of treatment. Summary data of % bee death per day show little difference in % bee death for the first 48 hours of treatment among treatment at all three temperatures. Only after 72 hours was the % bee death greater than 10%, with % bee death increasing with each subsequent day of treatment.

Although bee losses approach an unacceptable level after the third day of treatment (20%), the majority of mites had fallen by the end of the second day at 40°C (80-90%) and even at 35°C (>50%). The length of time required to cause significant mite drop is an important factor in the practicality of such treatments, in addition to potential bee losses. Heat treatment could potentially be used to control the mite population of a colony without causing significant bee mortality if considerations are made to minimize the length of the treatment, thereby reducing both bee losses and labor.

It is important to note that increased temperature did not necessarily kill mites. However, the temperature treatments did result in increased mite drop where mites were subsequently trapped on a sticky board. In a typical hive situation mites that fall from bees are able to crawl back up onto bees in the absence of sticky boards or other traps. The use of sticky boards or other traps such as recessed bottom boards probably decreases the amount of mite infestation by trapping mites that fall as a natural occurrence. This may help explain why recessed bottom boards may show potential in controlling mites, particularly during active brood rearing temperatures when much of the hive is kept at 35°C (Pettis and Shimanuki 1999).

Several factors may contribute to the tendency of mites to drop while under heat treatment. Mites have a higher surface area/volume ratio than bees and therefore dessicate faster. Temperatures over 38°C impair mite vitality, which may cause them to drop (Engels and Rosenkranz 1992). Heat also increases bee respiration and movement which might also contribute to increased mite drop (Harbo 2000).

Although heat treatment for control of varroa mites has been a common practice in Eastern Europe for many years, it poses the inherent problem of potentially killing bees, in addition to inducing mite drop. Typically, such procedures involve the use of dangerously high temperatures for short periods of time, necessitating either the transfer of bees from the hive into a heating chamber or the removal of brood frames. Because this is labor-intensive, it is not feasible to use such heat treatments in large apiaries.

We found that it may be possible to significantly decrease mite levels without exposing adult bees or bee brood to dangerously high temperatures by increasing hive temperatures in a controlled fashion over specific time periods. This would also reduce the amount of labor required to conduct such a treatment. The next step in this research is to test this process on entire bee colonies (adults and brood) in whole hive situations.

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An Evaluation of Far-eastern Russian Honey Bees and Other Methods for the Control of Tracheal Mites

by LILIA I. DE GUZMAN[1], THOMAS E. RINDERER[1], GARY T. DELATTE[1], J. ANTHONY STELZER[1], LORRAINE BEAMAN[1], and CHARLIE HARPER[2]

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Abstract

The effects of several honey bee management tools on tracheal mite (*Acarapis woodi*) populations were evaluated in southern Louisiana. Both domestic and ARS Primorsky honey bee colonies were evaluated after: 1) a single application of Beltsville formic acid (BFA) to hives with a solid bottom hoard, 2) a single application of BFA to hives with a screen bottom hoard, 3) no treatment with a screen hottom board, or 4) no treatment with a solid bottom hoard. Domestic colonies in hives with screen hottom hoards or hives with solid bottom boards maintained high levels of tracheal mite infestation throughout the experiment. However, in domestic colonies, the use of a combination of BFA and a screen bottom board reduced tracheal mite infestation levels by 47% after three months and maintained the lowered levels for an additional three months. The use of BFA and a solid bottom board resulted in a 37% reduction of tracheal mites after three months, but levels then increased to a damaging level by six months. Tracheal mite populations remained very low in Primorsky colonies, regardless of the management tools employed.

Several conclusions are apparent: 1) The ARS Primorsky honey bees tested exhibited strong resistance to tracheal mites and do not require treatment for tracheal mite control, 2) the commercial honey bee stock tested displayed high susceptibility to tracheal mites, 3) one summer treatment of BFA did not adequately control tracheal mite populations in highly susceptible honey bee stock, 4) BFA showed moderate tracheal mite control and may be useful in Integrated Pest Management (IPM) programs designed to control the most serions honey bee pests and diseases, and 5) while screen bottom boards did not suppress tracheal mite populations, they enhanced the effectiveness of BFA in tracheal mite control.

KEYWORDS: *Acarapis woodi*/tracheal mite/ARS Primorsky/ resistance/formic acid/screen/IPM/ far-eastern Russia/ USA

INTRODUCTION

Acarapis woodi is an internal parasitic mite that lives in the prothroracic tracheae of honey bees. High infestations of tracheal mites usually cause mortality of honey bee colonies. Colony losses are more apparent in harsh winter conditions in the northern region of the United States, but can also occur in less adverse seasons and weather conditions.

The control of tracheal mites remains an important concern in beekeeping. Chemical control options are limited. Only a few chemicals are known to be effective. Generally these chemicals vaporize in a hive and are delivered to the site of tracheal mite infestation by the bees' respiratory system. Several compounds of botanical origin are effective against tracheal mites (Cox et al. 1989, Calderone et al. 1991, Ellis and Baxendale 1997, Sammataro et al. 1998, Elzen et al. 2000). However, only menthol has regulatory approval for use in tracheal mite control in honey bee hives.

Formic acid is a naturally occurring compound, which also is reasonably effective in controlling tracheal mites. The effects of liquid formic acid on *Varroa destructor* and *A. woodi* have been studied extensively (Bracey and Fischer 1989, Hoppe et al. 1989, Fries 1991, Wilson et al. 1993, Calderone and Nasr 1999, Baxter et al. 2000, Calderone 2000). These studies showed efficacies of 50% to 80% on *V. destructor* and 87% to 99% efficacies on *A. woodi*. However, the hazardous effects of formic acid on both beekeepers and their honey bees remains a serious concern. This hazard is reduced when the formic acid is in a gel formulation. The Beltsville gel formulation of formic acid (BFA) is considered safer, may achieve as much as 70% efficacy for *V. destructor* control (Feldlaufer et al. 1997) and has regulatory approval for use in honey bee hives. Also, in cage tests lasting 8 days, BFA caused nearly 100% mortality of tracheal mites (Feldlaufer et al. 1997), although the effectiveness of BFA against tracheal mites in field conditions has not been reported. Like the liquid form of formic acid, BFA has also been found to have some drawbacks such as reducing drone production and adult drone survival (Guzman et al. 1999) and its use requires strict safety procedures to prevent harm to beckeepers. Nonetheless, it has a potential role to play in the control of honey bee mite parasites.

As an alternative to chemical controls, some researchers have investigated the potential for honey bee stocks to be bred which are resistant to tracheal mites (Gary et al. 1990, Milne et al. 1991, Rinderer et al. 1993, Danka et al. 1995, Williams et al. 1994, Lin et al. 1996, Guzman et al. 1998 a & b). These efforts have had substantial success and several stocks of honey bees which are resistant to tracheal mites are marketed to the beekeeping industry.

This experiment is part of a larger project of the Honey Bee Breeding and Genetics Physiology Laboratory to develop Integrated Pest Management (IPM) recommendations, the objective of which is to integrate effective control measures of the major pests and diseases of honey bee colonies. It is necessary in the construction of IPM plans to combine treatments that substantially enhance the effects of component treatments which target one or more pests. In this report we evaluate the effectiveness of *A. woodi* control treatments when combined with treatments

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