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

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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 board, 2) a single application of BFA to hives with a screen bottom board, 3) no treatment with a screen bottom board, or 4) no treatment with a solid bottom board. Domestic colonies in hives with screen bottom boards 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 serious 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 beekeepers. 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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designed for the simultaneous control of *V.destructor*. A companion report will provide similar information for the effectiveness of BFA for the control of *V. destructor*. The application of formic acid in July was not an ideal time for tracheal mite control, but was done primarily to control *Varroa* mites. Natural infestations of tracheal mites in the colonies in this experiment were monitored and results from the first year of the study are reported here.

## MATERIALS AND METHODS

Domestic honey bee colonies in Carencro, Louisiana were divided into five-frame nucleus colonies in April 2000. Island-mated ARS Primorsky queens and domestic queens (purchased from a commercial queen breeder) were randomly assigned and installed into the nuclei. The nucleus colonies were placed in a holding yard for two weeks until queens were accepted and the colonies were transferred into standard Langstroth hives in May 2000. At that time, modified screen bottom boards with attached 8-mesh hardware cloth screens similar to those described by Pettis and Shimanuki (1999) were installed for test groups designated as the screen group (n=12 for each stock), and the screen plus formic acid group (n=12 for each stock). A total of 96 colonies were relocated and used to form three apiaries of 32 colonies each. The colonies were set on 4-way pallets; each pallet had only one type of queen and one type of treatment. Infestations of tracheal mites in all colonies were determined in July 2000 prior to BFA treatment. The formic acid, and the formic acid and screen treatment groups received one treatment of one pack of formic acid gel on July 26, 2000, which was removed 11 weeks later. Mite counts were determined in October 2000, January and April 2001.

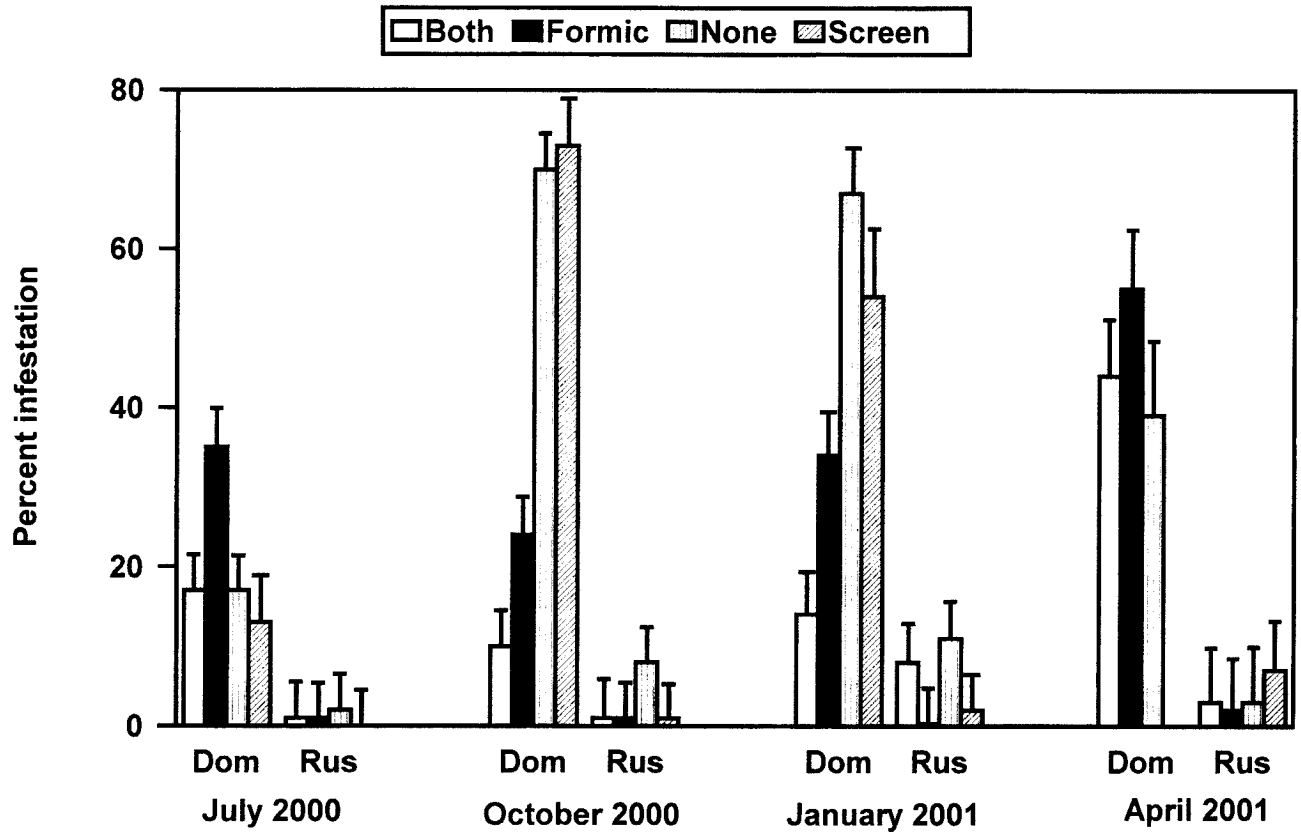
Tracheal mite prevalence (number of infested bees/total number of bees in sample) and mite intensity (number of mites/infested bee) were determined for each colony. Infestation parameters were determined by examining 30 worker bees per colony. Immature stages of mites with a "saggy" appearance were considered dead and thus were not included in the count. However, dead and live adult mites were not differentiated since bee samples were frozen for a few days before all dissections were completed. Adult bee populations were estimated as described by Burgett and Burikam (1985).

Data on mite prevalence, mean mite intensity and adult bee populations for July and October 2000 were analyzed using ANOVA of a factorial experiment in a Completely Randomized Design. In each site, a split-plot was employed where the main unit was honey bee type, colony was replication and sampling date was the subunit. Honey bee type, treatment type, sampling date, and apiary site were modeled as fixed effects using Proc Mixed. Colony within type, site and treatment were modeled as random effects. Data collected in January and April 2001 were analyzed separately due to substantial colony losses of domestic colonies. Degrees of freedom were estimated using the Kenward-Roger method. Before analyses, data for the prevalence were arcsine transformed and mite intensity were transformed using square root transformation. The Pearson correlation coefficient was used to test the relationship between the number of adult bees and tracheal mite prevalence. (SAS Institute 1997).

## RESULTS

**Prevalence.** Our results for July and October showed a significant interaction between bee type, treatment and sampling month ($P<0.0001$). In the domestic colonies, the untreated group significantly ($P<0.0001$) increased in the rate of infestation by 312% (from 17 to 70%) in October (Figure 1). Similar infestation was

**Figure 1. Prevalence of *A. woodi* in ARS Primorsky (Rus) and domestic (Dom) honey bee colonies as affected by different control methods. Both = one application of BFA with a screen bottom board, Formic = one application of BFA with a solid bottom board, None = no treatment with a solid bottom, Screen = no treatment with a screen bottom board.**



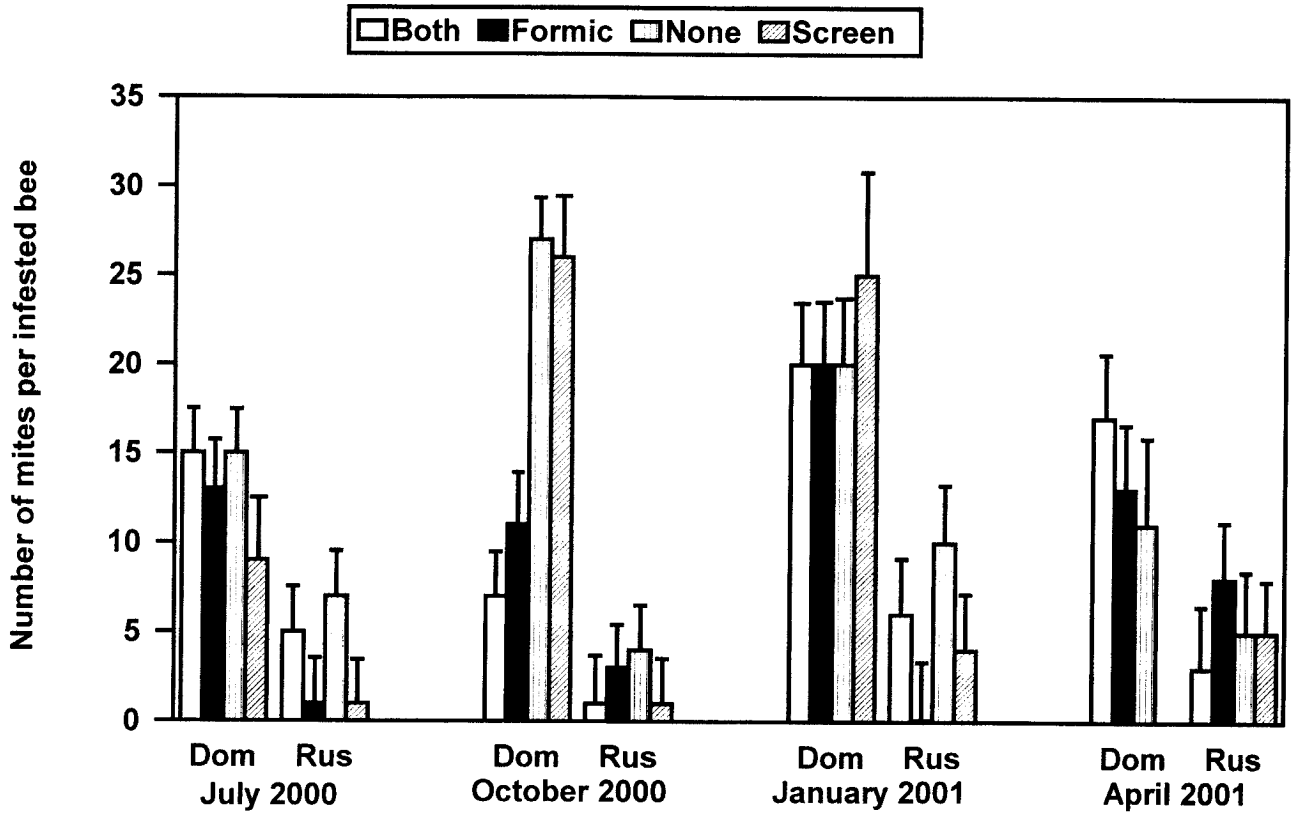
observed in January 2001. Due to the deaths of highly infested colonies, infestation in the untreated group decreased in April to a mean of 39%, which was still well above the economic threshold level of 25%. The colonies with screen bottom boards had equally significant ($P<0.0001$) increase of 462% (from 13 to 73%) in October. In this group, there was a slight decrease in average infestation observed in January when highly infested colonies died due to tracheal mite infestations. No domestic colony in the screen bottom board group survived to April. Formic acid did not significantly ($P=0.068$) reduce the infestation (from 35 to 24%) from July to October. Tracheal mite infestation in this group gradually increased through January with a peak of 55% observed in April. The combination of formic acid and screen bottom board decreased significantly ($P=0.0151$) from 17 to 10% in October, three months after treatment was applied. Tracheal mite infestation in this group remained low in January, but increased to 44% in April. For the Primorsky colonies, significantly low infestations were observed in both July and October, regardless of the treatment employed ($P=0.4168$). A numerical increase in infestation in the untreated group was observed in October (from 2 to 8%), but was not statistically different ($P=0.1376$) from the infestation in July. Infestation increased to 11% in January and declined to an average of 4% in April when the only three highly (33, 47 and 53%) infested Primorsky colonies died due to cold weather exposure when hive covers were blown off during a rainstorm.

In all sampling periods, the Primorsky colonies had significantly ($P<0.0001$) lower infestation levels than the domestic colonies. At the end of the experiment, all surviving Primorsky colonies had tracheal mite infestations that ranged from 0-17% (Table 1). No significant ($P=0.6895$) influence by apiary sites was observed.

**Mite Intensity.** Figure 2 shows the effects of different control methods in the intensity of tracheal mites in both domestic and Primorsky colonies. A significant ($P<0.0005$) interaction between type, treatment and sampling month was also observed after three months of observation. In the domestic colonies, infested bees from all treatment groups had about 11-15 mites in July 2000. Eleven weeks after treatment, mite intensity in the formic acid group decreased by 15% (from 13 to 11 mites), increased sharply to 20 mites in January and later declined to 13 mites per infested bee in April. Mite intensities were lower in April and July when brood was present and new hosts were emerging in the colonies. The formic and screen combination group had a 53% (from 15 to 7 mites) decline in mite intensity in October and then rose to a peak (20 mites) in January. The mite intensity in the untreated domestic and screen bottom groups in October increased significantly ($P<0.0001$) from 15 to 27 and 11 to 28 mites per infested bee, respectively. Mite intensities in these groups decreased to 20 mites in the untreated and 25 mites in the screen group in January. The Primorsky colonies had lower mite intensities than the domestic colonies throughout the experimental period. The highest intensity (10 mites) among the Primosky colonies was observed in the untreated group in January when very few young newly-emerged hosts were present in the colonies. Apiary sites did not influence ($P=0.336$) mite intensity.

**Adult bee populations.** Our results showed a significant ($P=0.0132$) interaction between type and sampling month. In July, domestic colonies had significantly ($P<0.0001$) more adult bees than the Primorsky colonies, since Primorsky colonies tend to reduce their populations during nectar and pollen dearths. Both types had similar ($P=0.1202$) numbers of adult bees in October during the Autumn flow. No correlation was detected between the

**Figure 2. Intensity of *A. woodi* in ARS Primosky (Rus) and domestic (Dom) honey bee colonies as affected by different control methods. Both = one application of BFA with a screen bottom board, Formic = one application of BFA with a solid bottom board, None = no treatment with a solid bottom, Screen = no treatment with a screen bottom board.**



number of adult bees and tracheal mite prevalence in either domestic (r = -0.2008, $P$=0.0653) or Primorsky (r = -0.02787, $P$=0.7897) honey bee colonies.

At the end of the experiment in April, only six domestic colonies had good bee populations with sufficient brood to make up at least one new colony with 3-4 brood frames and two frames with honey and pollen from each colony (Table 1). One of the 12 undivided domestic colonies was very weak and needed additional frames of brood and bees. For the Primorsky honey bees, 19 strong colonies were divided to make new splits. Each of the sixteen undivided Primorsky colonies was strong enough to receive a new queen without requiring additional brood or bees.

**Dead colonies**. Based on experience, highly infested colonies will not survive through the winter. In October, we predicted that 10 domestic colonies would die in the winter due to their high infestations (73-100%) of tracheal mites. Some of these colonies had several bees crawling in front of the hives. Crawlers from two of these colonies showed 70 and 100% tracheal mite infestations. Crawling of infested bees may have been aggravated by relatively cool (9-19°C) weather in October 2000. In January and April 2001, an additional 16 domestic colonies died from mite infestations (Table 1). This high mortality of domestic colonies might have been accelerated by the presence of *Varroa* mites in the colonies. All dead domestic colonies except three (0, 7 and 23%) had high tracheal mite infestations that ranged from 37-100%. The five dead Primorsky colonies had lids blown off by strong winds and were then exposed to cold temperature and rains between January and March 2001. The last tracheal mite infestations recorded from these Primorsky colonies were 7, 10, 33, 47 and 53%. The actual cause of death of the three Primorsky colonies with more than 33% infestation is unclear. Whether or not the colonies would have tolerated such infestations cannot be determined since these were the only Primorsky colonies with infestations higher than 20% throughout the experiment.

## Discussion

In the Primorsky honey bee colonies, the strong resistance to tracheal mite infestation produced an overriding effect. Regardless of treatment, the Primorsky colonies had very few tracheal mites. In apiaries with some domestic colonies having infestation rates approaching 100% during summer in Louisiana, the highest infestations in the Primorsky colonies averaged about 11%, a level far below the economic threshold of 25% reported by Eischen (1987). The Primorsky colonies' ability to maintain infestations at low levels was also apparent from the infestation rates observed in July, three months after the introduction of queens into the colonies and the installation of screen bottom boards in the screen, and screen and formic acid groups. Although queens were assigned to colonies randomly, the Primorsky colonies had far fewer mites than did the domestic colonies. These observations are consistent with our earlier studies showing the same ability of Primorsky honey bee colonies to maintain low levels of tracheal mite infestations (Guzman et al. 2001a & b, In Press). Clearly, the best approach to including tracheal mite control in an overall IPM plan for honey bees is the use of stock that is strongly resistant to tracheal mites.

Tracheal mite resistant stocks are available to the U. S. beekeeping industry. ARS Primorsky honey bees, Yugoslavian honey bees, and Buckfast honey bees have all been specifically documented to be resistant to tracheal mites (Rinderer, et al.1993, Williams et al. 1994, Danka et al. 1995, Lin et al. 1996, Guzman et al. 1998 a & b, Guzman et al. 2001 a & b, In Press). Additionally, procedures for selecting tracheal mite resistant stock are available and some queen breeders have resistant breeder queens in their own stock (Danka and Villa 2000). However, tracheal mite resistance is not present in all commercially available stocks: some have no apparent resistance and some have only moderate levels of resistance.

The domestic stock used in this experiment showed strong susceptibility to tracheal mites. The development of very high levels of tracheal mite populations in honey bee colonies in summer and early autumn in Louisiana is not typical. However, the susceptibility of the domestic stock did provide a framework of potentially high mite levels in which to evaluate the management treatments. The domestic colonies receiving a single application of BFA had about 1/3 the prevalence of mites when compared to similar untreated colonies three months after treatment application. The prevalence of tracheal mites in October after the BFA treatment in July was still high (24%), probably owing to the strong susceptibility of the stock. Infestation levels higher than 20% were reported to have caused deaths of honey bee colonies in New York (Otis and Scott-Dupree 1992). Tracheal mite infestation in the formic acid group increased to 34 and 55% in January and April, respectively, which suggests that one application of formic acid was not sufficient to protect the susceptible colonies from tracheal mite parasitism. An additional treatment of BFA in Autumn may have prevented the increase in infestation observed in January and April.

The domestic colonies receiving a single application of BFA and having screen bottom boards had an even smaller prevalence of mites in October (about 1/5) and January (about 1/2) when compared to similar untreated colonies. The screen bottom boards themselves had no apparent influence on mite numbers. However, the combination of screen bottom boards and BFA brought the prevalence of mites to an average below the economic threshold for six months. The presence of a screen bottom board enhanced the effectiveness of the BFA treatment in some unknown manner, perhaps by facilitating a good dispersal of formic acid vapors throughout the hive. This result is contrary to the intuitive expectation that the large screen bottom board would be expected to cause a rapid reduction in the concentration of formic acid vapor to ineffective levels.

The effects of tracheal mites in the southern United States were thought to be of minimal consequence to the honey bee colonies

**Table 1. Numbers of colonies infested with tracheal mites, surviving colonies for each treatment, dead colonies that provided colony divisions (splits) at the end of the experiment in April 2001.**

| | Domestic | Primorsky |
|---|---|---|
| Infestation range: | | |
| 0 | 0 | 19 |
| 1-10 | 4 | 11 |
| 11-20 | 2 | 5 |
| 21-50 | 3 | 0 |
| >50 | 9 | 0 |
| Treatments: | | |
| 1. One application of BFA with a solid bottom board* | 7 alive<br>4 dead<br>(2 SS****) | 9 alive<br>0 dead<br>(2 SS) |
| 2. One application of BFA with a screen bottom board | 7 alive<br>3 dead<br>(2 SS) | 8 alive<br>2 dead<br>(2 SS) |
| 3. No treatment with a screen bottom board | 0 alive<br>11 dead<br>(1 SS) | 10 alive<br>0 dead<br>(1 queenless, 1 SS) |
| 4. No treatment with a solid bottom board | 4 alive<br>8 dead<br>(0 SS) | 8 alive<br>3 dead<br>(1 SS) |
| No. of surviving colonies | 18 | 35 |
| No. of dead colonies | 26** | 5*** |
| No. of colonies with no division made | 12 | 16 |
| No. of colonies with divisions made | 6 | 19 |

* 13 Domestic and11 Primorsky colonies were treated.
** 10 colonies culled due to high tracheal mite infestations in October 2000.
***lids blown off, colonies were exposed to cold temperature and rains between Jan. and March 2001.
****SS = supersedure, colonies with SS queens were dropped from the experiment.

due to hot weather in summer. The death of more than half of our domestic colonies due to high tracheal mite infestations and the inability of surviving colonies to build up in spring clearly shows the potential severity of tracheal mite parasitism in Louisiana. Small populations of bees in springtime colonies may be due to the increased death of highly infested bees. Our observation corroborates the findings of Eischen (1987) showing a strong association between tracheal mite infestation and colony size. High infestations of domestic colonies in Louisiana also have been reported in our earlier studies with different domestic stocks (Guzman et al. 2001 a & b).

**Conclusions:**

1. The strong resistance of Primorsky honey bees to tracheal mites in itself was sufficient to minimize tracheal mite damage.
2. One treatment of BFA in July reduced the prevalence of tracheal mites, but under conditions of this test, the reduction was insufficient for protecting a highly susceptible honey bee stock. A second BFA treatment in Autumn may be needed to further reduce infestations.
3. Screen bottom boards alone neither increased nor decreased the prevalence of tracheal mites.
4. Screen bottom boards enhanced the effectiveness of BFA.

**Recommendations:**

1. Stock with at least some resistance to tracheal mites and preferably strong resistance to tracheal mites is an essential component of an IPM program designed to control major honey bee pests and diseases.
2. Where moderately resistant stock is desirable for some other reason, or perhaps in areas highly conducive to the development of tracheal mite infestations, single treatments of BFA will reduce tracheal mite numbers and screen bottom boards will enhance the effectiveness of BFA.

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