ATTTA
Atlantic Tech Transfer Team
for Apiculture

Report - Initial Findings on Miticide Efficacy in the Maritimes

In 2017, the Atlantic Tech Transfer Team for Apiculture (ATTTA) conducted two separate trials in which the efficacy of miticides used to control populations of the ectoparasitic varroa mite (*Varroa destructor* Anderson and Trueman) afflicting colonies of western honey bees (*Apis mellifera* L.) were assessed. A spring field trial in Northern Nova Scotia indicates amitraz, fluvalinate, and thymol oil to be effective varroa mite treatments. A summer resistance monitoring trial in Nova Scotia suggests resistance in populations of varroa mites to both amitraz and the newly registered flumethrin to be unlikely.

Introduction

Varroa mites are the most serious pest afflicting western honey bees and have been reported to be the main culprit for overwintering colony mortality in Ontario (Guzmán-Novoa et al, 2010). If left unchecked, populations of varroa can seriously compromise entire colonies through direct feeding and the vectoring of viruses, potentially to the point of colony failure and collapse.

Several products are registered in Canada to control populations of varroa mites, including synthetic pesticides and naturally occurring organic acids. These products are commercially available and are regularly used by Maritime Canadian beekeepers. The frequent and widespread application of synthetic pesticides raises concerns for reduced efficacy through the possible development of resistance. Such circumstances are documented in various parts of the world where honey bees are managed commercially, including Canada (Currie et al, 2010). The development of resistance to one product prompts the registration of another. Checkmite+® (Bayer) and Apistan® (Wellmark International) are examples of synthetic miticide products to which resistance has been observed in varroa mites in Canada. See ATTTA's *Condensed Report on Miticide Resistance in Atlantic Canada* for more information.

The current industry standard product for controlling varroa in the Maritimes is Apivar® (Vetò-Pharma), whose active ingredient is the formamidine insecticide amitraz. Beekeepers in New Brunswick, Nova Scotia, and Prince Edward Island report using Apivar® both as a spring and fall treatment (although most likely only apply the product to their hives once a year) (Ferland et al, 2017). It is unknown whether resistance to amitraz exists in Canada but resistance has been reported in The United States as early as 1999 (Elzen et al, 1999). ATTTA sought in 2017 to help determine if this product is still effective in the Maritimes through a pair of trials.

Trial 1: In-hive testing

Methods

In late April, 44 hives from three different apiaries operated by the same beekeeper in Northern Nova Scotia were sampled for their varroa mite levels. Apivar®, Apistan®, and one commercial 'natural' miticide alternative – Thymovar® (BioVet AG), a thymol essential oil based product – were each placed in 12 – 14 of the 44 hives following label recommended application rates (n = 14, 12, and 12 for Apivar®, Apistan®, and Thymovar, respectively). The treatment period for all three products is six weeks (i.e. 42 days), however, strips of Thymovar® are recommended to be removed and replaced after three weeks, while the other two products remain in the hives for the entire six weeks. This spring treatment period preceded the transportation of the hives to commercially managed fields of blooming lowbush blueberry (*Vaccinium angustifolium*). Many honey bee hives in the Maritimes are managed for their annual lowbush blueberry pollination service. No honey supers were in place at this time and hives were not fed sugar syrup supplements.

Growing Forward 2 | Nova Scotia | New Brunswick / Nouveau Brunswick | Prince Edward Island | Canada

Funders and Contributors:

Bleuets NB Blueberries
New Brunswick Beekeepers Association Inc.
Nova Scotia Beekeepers' Association
Wild Blueberry Producers' Association of Nova Scotia
Prince Edward Island Wild Blueberry Growers Association
PEI Beekeepers' Association
Oxford Frozen Foods
Jasper Wyman and Son

Prior to the initiation of treatment, mite levels were assessed with an alcohol wash. After the recommended treatment period, the miticides were removed from the hives in early June and mite levels were assessed again. A small control group of four (n = 4) hives in one apiary was included to act as a baseline comparison of potential mite levels if left untreated over the spring treatment period. The control group was kept relatively small to reduce the risk of mite populations drastically increasing during the treatment period which could potentially compromise the beekeeper's operation.

In addition to assessing pre and post-treatment varroa levels, the relative strength of each colony was assessed by counting the number of "seams of bees", defined as the spaces in between frames within a hive mostly or entirely occupied by worker honey bees. For instance, a weak one-storey colony may only have two or three seams of bees while a strong colony may have nine or ten. Furthermore, colonies were visually inspected for symptoms of deformed wing virus, sacbrood bee virus, and Israeli acute paralysis virus, all of which are known to be vectored by varroa mites (Pernal et al, 2013).

Results and Discussion

Although many of the hives' varroa mite levels were relatively low before treatment (i.e. less than the spring economic threshold of 2 mites per 100 bees as described by Eccles et al, 2013a in Ontario), statistically significant mite reduction was still achieved in this trial. The average mite levels for all three miticides were significantly reduced after treatment at the 95% confidence level (p = 0.037, 0.018, and 0.045 for Apivar®, Apistan®, and Thymovar®, respectively) (Figure 1). Average mite levels post-treatment for each miticide also differed significantly than the average mite level of the control group at the 95% confidence level (p = 0.005) (figure 1). The treatment effect between the three miticides however, did not differ significantly at the 95% confidence level (p = 0.330) (Figure 1).

Most of the colonies included in the trial increased in size and strength over the treatment period, while some decreased. Overall, the colonies in the Apivar®, Apistan®, Thymovar®, and control groups increased by an average net gain of 1.50, 3.58, 3.15, and 4.38 seams of bees, respectively (Figure 2). Although the highest average colony size increased occurred in the control group, no significant difference in net colony size gain was observed among the treatment and control groups (p = 0.656) (Figure 2).

No evidence of deformed wing virus, sacbrood virus, or Israeli acute paralysis virus was observed in any colony.

Despite documented resistance to fluvalinate in Canada (Currie et al, 2010), Apistan® was effective at controlling varroa mites in the hives included in this trial. This is likely due to a gap of several years since the product was last used in the operation. The exact date of the last application of Apistan® is unknown but was suggested to be at least several years by the operating beekeeper. Of notable interest is the fact that Thymovar® controlled mites as effectively as the other two synthetic miticides, particularly considering many of the spring days during the trial were cool and below the optimal outdoor temperature for treatment recommended by the manufacturers (i.e. 13 °C – 30 °C, reported by Eccles et al, 2013b). As expected, Apivar® was effective and continues to be the industry standard in Canada for varroa mite treatments. The data from this trial reinforces the importance of treating for varroa mites in the spring, or at least monitoring for them and ensuring their levels are below the spring treatment threshold.

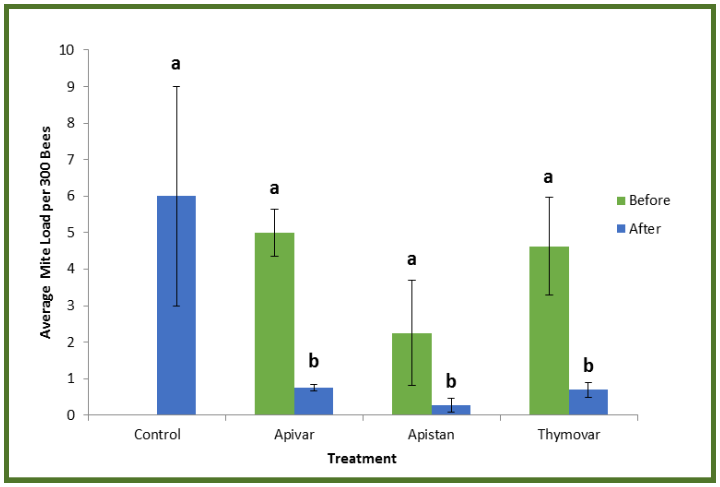

Figure 1: Comparison of varroa mite levels before and after treatment with Apistan® (fluvalinate), Apivar® (amitraz), and Thymovar® (thymol) (Note: bars labeled with different letters are significantly different from each other.)

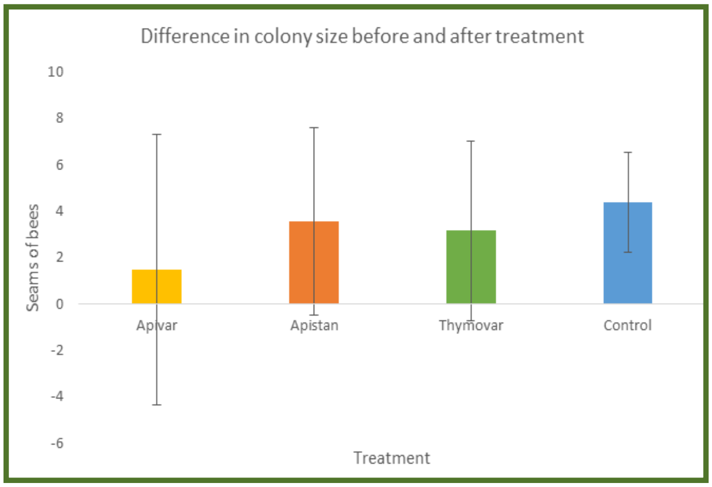

Figure 2: Net colony change in colony size over the course of the treatment period for each treatment group.

Trial 2: Resistance Monitoring

In late 2016, Health Canada's Pest Management Regulatory Agency approved the registration of Bayvarol® (Bayer), a synthetic insecticide based product whose active ingredient is flumethrin, to control varroa mites in honey bee hives. Flumethrin, like fluvalinate which is the active ingredient in Apistan®, is a class 3A pyrethroid insecticide. Although fluvalinate has never been registered before in Canada to control varroa mites, there are concerns over the possibility of cross-resistance caused by historical usage of fluvalinate because the two insecticides are in the same class. ATTTA sought in 2017 to monitor for evidence of any such cross-resistance existing in Maritime Canada.

Methods

In the late summer of 2017, ATTTA collected samples of live bees from various colonies around the Maritimes and placed them in special incubation kits. Each kit contained a section of a strip of either Apivar®, Bayvarol®, or cardboard as a control treatment. Samples of approximately 300 bees were each placed into one sampling kit each and incubated for 3 – 6 hours. Following the incubation period, varroa mites killed by the miticide strips were counted, and the bees were washed with alcohol to dislodge the remaining mites that were not killed by the miticides. A ratio of mites whose mortality was induced by a particular miticide vs how many survived the treatment was then deduced for each bee sample and the average percent efficacy for the miticide was calculated across all the bee samples taken. Samples in which $< 50\%$ of mites are killed indicate potential resistance.

Some mite mortality was observed in the control samples during the incubation period despite no miticide strip present in the kits. The mortality observed in the control samples was used to mathematically adjust the percent efficacy observed in the Apivar® and Bayvarol® samples using the Abbott's correction formula to account for any mortality that was not induced by the miticides themselves (Abbott, 1925).

In total, 53 valid samples (i.e. > 5 mites per 300 bees) were obtained and incubated. From Nova Scotia, 36 valid samples (13 control, 13 Apivar®, and 10 Bayvarol®) were collected. From New Brunswick, nine valid samples (1 control, 4 amitraz, and 4 flumethrin) were collected. From PEI, 8 valid samples (2 control, 3 Apivar®, and 3 Bayvarol®) were collected. These samples were collected from four apiaries in Nova Scotia, and one apiary each from New Brunswick and Prince Edward Island.

Results and Discussion

On average, Bayvarol® killed approximately 92% ($n = 17$) of the varroa mites sampled and Apivar® killed approximately 99% ($n = 20$) (Figure 3). Both of the aforementioned averages were adjusted using Abbott's correction of approximately 19% which was the average mite drop in the control samples ($n = 16$). Significantly more mortality was observed in both treated samples than the control samples at the 95% confidence level ($p < 0.0001$) (Figure 3). The mortality observed with Apivar® however, was significantly higher than Bayvarol® at the 95% confidence level ($p = 0.004$). Despite the difference in treatment efficacy between both products, both Apivar® and Bayvarol® killed $> 50\%$ of mites in all samples, indicating resistance to either of these products to be unlikely in the populations of mites sampled.

In addition to the valid samples taken, numerous others were collected from New Brunswick and Prince Edward Island so that a roughly equal proportion was sampled from each Maritime province. Unfortunately, most of the samples taken from these two provinces did not have a sufficient number of varroa mites in them to be considered valid and are thus excluded from this report. This initial sampling in 2017 however, suggests Apivar® and Bayvarol® are effective varroa mite treatment products, and lays the groundwork for a more comprehensive assessment of efficacy across the Maritimes and survey for any possible resistance.

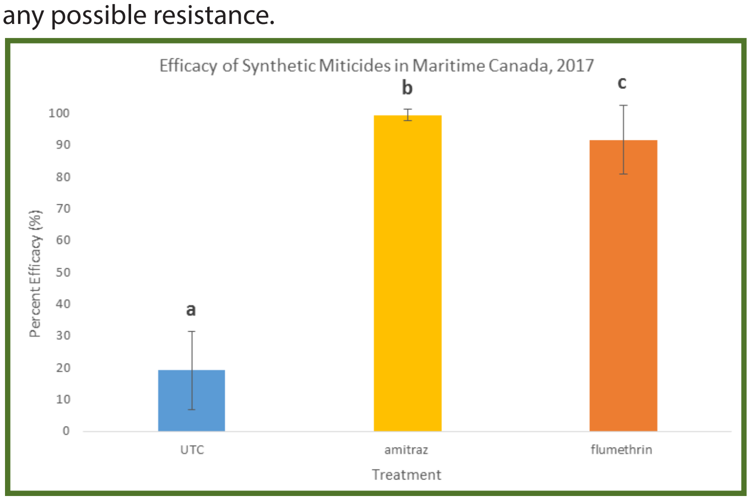

Figure 3: Comparison of efficacy of Apivar® (amitraz) and Bayvarol® (flumethrin) in incubation bioassay vs an untreated control (UTC) (Note: bars labeled with different letters are significantly different from each other.)

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October 2017