Mite-Away Quick Strip™ Mid Honey Flow Efficacy Trial

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Monitoring phoretic Varroa destructor levels after the application of a strip formulated acaricide (Mite-Away Quick Strip™[3]), targeting the male varroa. Formic acid, in a saccharide gel matrix, is the active ingredient.

Abstract

Traditional approaches to controlling the parasitic mite *Varroa destructor* in honeybee colonies have focused on targeting the female mite when phoretic (outside the brood cells, on the adult bees). NOD Apiary Products Ltd. (NOD) decided to shift its focus to killing the male varroa. The hypothesis is that, if the males are killed prior to mating, the female varroa population will remain low for an extended period of time.

Working with formic acid as the active ingredient, NOD developed a single application, strip formulated, "sticky" formic acid vapour release product (Mite-Away Quick Strips™), using saccharides. Mite-Away Quick Strips™ are inserted into the brood rearing area of the colony. The air movement response of the honeybee colony to formic acid vapours is utilized to drive the molecule under the cap, to kill the male varroa.

Determining the overall efficacy is a challenge in this model of varroa control. The traditional methods of determining efficacy (positive control comparison of phoretic female varroa kill) will not give a fully accurate efficacy picture. Mite-Away Quick Strips™ are designed for kill under the cap, so looking under the cap was on option. An alternative is an extended trial that monitors the trend of phoretic varroa mites using alcohol wash or a similar method. In 2009, NOD worked with researchers using all three methods. The study presented is a summertime, seven-week post-application, phoretic mite population trend study. Colony health, queen health, and the formic acid levels in the honey in honey supers, were also tracked.

Key words: varroa, male varroa, formic acid, Mite-Away Quick Strip™

The Male Varroa as a Target

The male varroa only live within the capped brood cell. Unlike the females, this makes them a non-moving target in a known location. The male varroa do not develop the hard outer shell of the females, so they should be more susceptible to formic acid vapours.

Varroa Sex

The foundress *Varroa destructor* mite lays her first egg 60-72 hours after the cell is capped (2.5 days at the earliest) and it is a male. The first female egg is laid 30 hours after that (3.75+ days after capping). It takes 5-6 days for the female to become sexually mature (9+ days after the cell is capped). The male produces sperm packets, which he takes into his mouth and places in the vagina of the most recently sexually mature female. Multiple matings are required over 30 hours to ensure full fertility (10 + days after the cell is sealed). In the reproduction of varroa, the second female varroa egg laid has a very low mating success rate in worker brood, high success rate in drone brood.

If the male varroa is killed at any time within 10 to 11 days of the cell being capped over, their reproductive capability has been impaired, even if the female sisters survive.

The Mite-Away Quick Strip™ is applied in the brood rearing area of the hive. An element of the Mite-Away Quick Strip™ treatment is the bee's natural ventilation response to the relatively high levels of formic acid vapours present immediately upon application. Enough formic acid vapours are able to penetrate the brood cell cappings if there is adequate air movement created in the brood area to drive the vapours through the brood cappings, into the cells. Therefore, a minimum colony size is a consideration.

Mite-Away Quick Strip™ Development.

NOD has been working with formic acid, as an acaricide, since 1997. Unlike conventional chemicals, no resistance is expected to develop to formic acid, and it is naturally occurring in honey.

To produce a biopesticide, to support the organic nature of formic acid, various saccharide based gel formulations were developed in NOD's laboratory. The goal was to have an effective rate of vapour release spanning three days. Unlike conventional pesticides, this product is designed to work with the bees; the colonies are not "passive recipients": The colony's drive to maintain temperature and humidity (homeostasis) in the brood rearing area is a factor in the product's effectiveness. The strip product was designed to fit in the bee-space, laid flat on frame top-bars.

After achieving success in incubator trials, field trials were conducted in 2009.

Materials and Methods

Apiary and Colony Selection

On July 6, 2009 (Day -4), in 2 bee yards, managed by David VanderDussen[4] to be "varroa farms", 21 colonies were identified as being queen-right, having healthy brood patterns, good size clusters for honey production, and having phoretic varroa loads at least double the recommended levels for the end of June Economic Threshold Level (ETL)[5]. Phoretic varroa loads were determined by using the alcohol wash method (approximately 300 adult bees per sample, taken from the brood rearing area). These colonies were allocated into three groups of seven; by location, brood chamber size, and mite loads. In the Control group, 6 of the 7 colonies were 2-brood chambers; each treatment group was 3 single brood chamber hives and 4 two-brood chamber hives. Other colonies in the yards were left untreated, providing additional varroa pressure, as most, if not all of the feral colonies in the area are gone.

The hives had been supered for the honey flow in June. Supers were left on during the treatment, and were added to as needed through the trial period.

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5 **July 1st varroa threshold is 1%. September 1st varroa threshold is 3% (Currie, R., University of Manitoba, Canadian Association of Professional Apiculturists published recommendations, Updated May 26, 2008) http://www.capabees.com/main/files/pdf/varroathreshold.pdf**

Figure 1. (l) Queen just after MAQS treatment, (r) brood combs 6-weeks after treatment.

Formic acid levels were monitored in 10 hives in the first 22 days of the trial. From each hive 5 samples were taken: At time of treatment (Day 0), Day+3, Day+7, Day+14, and Day+21. The honey samples were taken from the honey supers, each sample a composite from 5 points in the nearest super to the product application. Analysis was performed by Chemisar Laboratories Inc.[6]

Mite-Away Quick Strip™ Application

On July 10, 2009 (Day 0) Mite-Away Quick Strips™ were applied to the treatment groups. Group-1, as the Control, did not receive any treatment, but hives were opened and had honey samples taken, following the same process as the treated groups. Group-2 colonies each received a 200-gram dose, Group-3 colonies received 300-gram doses. For single brood chamber colonies, the strips were laid across the width of the hive body, on the top bars, spacing them apart for easy bee movement and air circulation around them. The queen excluder was replaced, set on above the strips. In 2-story colonies the strips were placed between the brood chambers, laid across the frames in the same manner.

Temperatures

Ambient temperatures in the first three days, the critical time of the treatment, are illustrated in Chart 3. Highs and lows ranged from 26.7º C (80ºF) to 11.4º C (52.5ºF).

Results

Efficacy

At the Day+3, the Day+14, and the Day+21 marks, in the treated colonies, the colonies treated at 200-gram doses had phoretic varroa load reductions of 64.4%, 75.7%, and 76.2%; the ones treated at 300-gram doses showed a reduction of 95.5% at both the Day +3 and the Day+ 14 marks, 96.2% at the Day+21 mark. See Chart 1.

The phoretic varroa to bee ratio trends, over the full 7-weeks of the trial, are illustrated in Chart 2.

Queen health

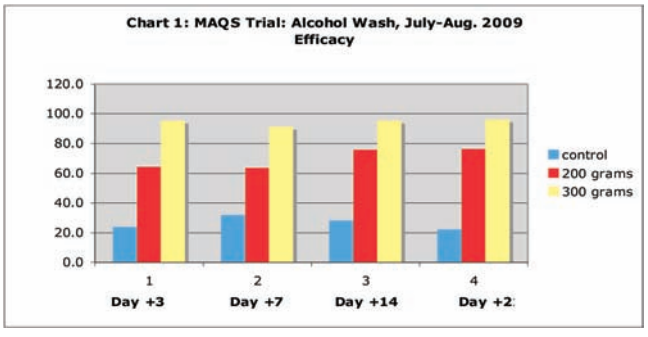
There was no sign of negative effects on the queens due to treatment. Egg laying continued throughout the 7-Day treatment period. In treated colonies, some damage to brood was observed at the Day+3 mark, but colonies recovered quickly. After Day+3 no additional damage was observed. Brood patterns were exceptionally solid and healthy in the treated colonies (see Figure 1); they became spotty, with signs of parasitic mite syndrome (PMS), in the Control colonies.

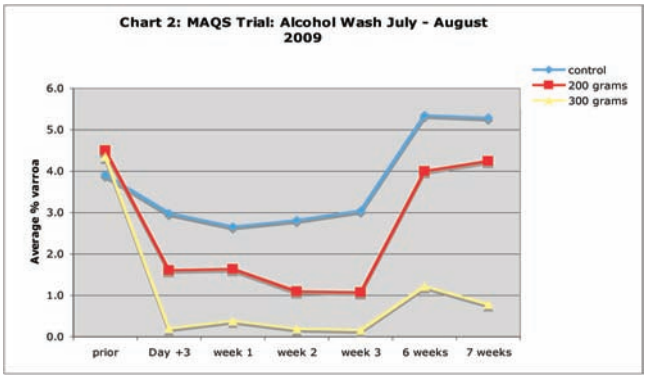
Colony health

Overall, colony health remained in good shape in all groups throughout the trial period. One Control colony became queenless just after the trial started and did not become queen-right by the end of the trial. Its varroa load level data was culled from the efficacy data set. One colony in the 300-gram group became a drone layer at some point after the Day+21 colony exam, so the Weeks 6 and 7 data on that hive was culled.

Formic acid levels in honey

Over the 22 days that the formic acid levels were monitored, formic acid levels in untreated colonies ranged from a low of 402 parts per





[6] **Chemisar Laboaratories Inc., 24 Corporate Court, Guelph, Ontario, Canada N1G 5G5 www.chemisar.com**

million (ppm) to a high of 2,097 ppm. In treated colonies the formic acid levels ranged from a low of 585 ppm to a high of 1,851 ppm. See Chart 4.

Discussion

The trend over the 7-weeks of the trial was an initial drop in phoretic varroa loads in all groups, with significant variation between groups by Day+3. The control colonies did exhibit the expected reduction in varroa-to-bee ratios that can occur in early summer, as the adult bee population growth rate exceeds the varroa growth rate. This is reversed in August. A similar pattern was observed in the 200-gram treatment group.

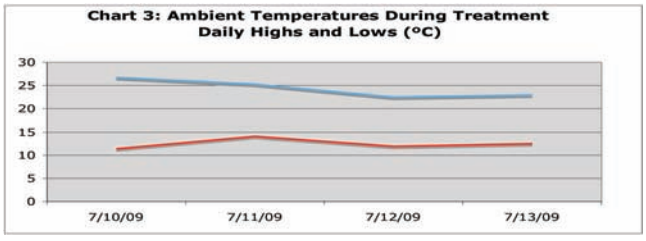
In the 300-gram treatment group varroa levels dropped to near zero, and remained low, even with high environmental varroa pressure present. This indicates that the treatment impacted the varroa population's reproductive capability.

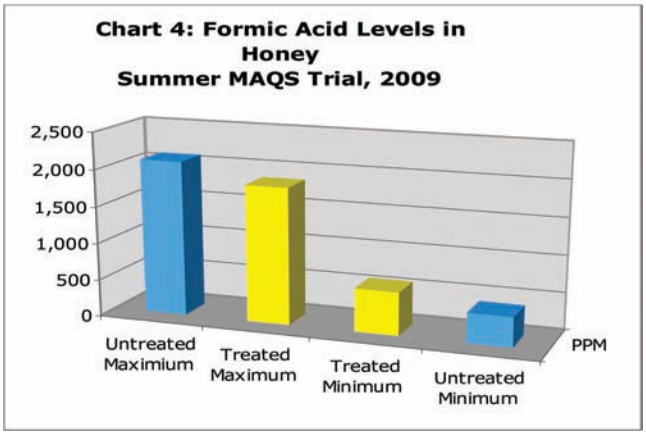
It should be noted that all the colonies in the trial were at least double the recommended varroa loads prior to the Mite-Away Quick Strip™ application, so the colonies were already under parasitic mite stress at the time of acaricide application. Environmental varroa pressure was maintained throughout the trial, making it similar to the migratory mingling, or "poor neighbor practices", re-infestation situations that can occur. Even under these stresses, the three-day treatment, at the 300-gram dose, was effective in bringing and keeping varroa loads under control for an extended period of time during a honey flow with supers on.

From an Integrated Pest Management (IPM) perspective, in the Control group, varroa loads stayed above threshold throughout the trial period, in the 200-gram colonies the loads had dropped to threshold but were again above threshold by the end of the trial. In the 300-gram treatment group, the drop in the phoretic mite load, especially for a vapour release product applied with supers on, was exceptional. A follow-up treatment in the early fall was unlikely to be required, especially if all colonies in the yards had received the 300-gram treatment, which would have greatly reduced environmental pressure. However, varroa levels should be monitored in late summer to ensure the winter cluster bees are adequately protected.

Colony health and queen health did not appear to be negatively affected by the treatment. Queens continued to lay well through the treatment period and to the end of the trial.

Formic acid is a component of honey, not an impurity. Therefore, the concern is not residues, but levels. The formic acid levels in the honey in the treated hives remained within what was determined to be naturally occurring. No withdrawal period is required.





Conclusion

One treatment, applied early summer on all colonies in a location, at the 300-gram dose application rate, was highly effective in knocking down varroa mite populations, and keeping them down, through the key harvestable honey production period. Indeed, in Northern climates, one treatment may provide sufficient control to keep varroa mite loads below treatment thresholds for one year, under the conditions tested. Two 300-gram treatments annually, one applied early summer and one applied later in the summer or early fall, should provide excellent control of varroa. Following an IPM program is recommended, to determine if an early-fall follow-up treatment is required.

Overall, neither queen nor colony health appears to be negatively impacted by the treatment. Formic acid levels tracked in the honey supers remained within what was naturally occurring, so no withdrawal period is required. Along with there being no chemical residues risk to the honey crop or wax, Mite-Away Quick Strips™ are an excellent new tool for controlling *Varroa destructor*, during honey flows.

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Figure 2. MAQS inserted between brood chambers during honey flow. When packed commercially, application will be two strips. Applicators must wear chemical gloves. Note the alcohol wash collection jars on the hives behind.