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### Colony Collapse Disorder

This is one way to bring our profession into the limelight, but not the best way. In case you have been living in a cave recently, commercial beekeepers (and some others) around the country have been watching their populous colonies dwindle right down to few or no bees in days. This started in the summer and continues to this day. The empty hives have honey, some have stored pollens, and sometimes significant patches of brood in them. The neglected brood appears to have every disease that we can recognize and other conditions seen only when brood suddenly is no longer is cared for, gets chilled, dies, and decomposes.

Besides all the media attention, honey bee researchers from the USDA bee lab in Beltsville, MD, the Department of Entomology and cooperating scientists from Penn State University, and researchers from the University of Montana are involved. They have scurried around the country taking samples of bees and combs (from collapsing and seemingly healthy colonies) and asking questions of each affected beekeeper. The last samples were taken from southcentralCalifornia last week, and now there will be thousands of hours of laboratory tests

conducted to see if there is a common, negative thread to this problem.

Dr. Jerry Bromenshenk from the University of Montana is conducting a Web survey of beekeepers having this problem and those who don't, to try to determine commonalities. You can do Jerry a big favor by going to the following Web site and entering data from your operation, good or bad: www.beesurvey.com.

One of the suspected culprits in these colony losses is the recently named species of what we used to call a microsporidian, but now they tell us it is a type of fungus, *Nosema ceranae*. That organism was named for its presence in *Apis cerana*. While we assume it originated in *A. cerana*, it was not found until after we introduced EHBs into Asia, so perhaps it moved over from EHBs.

We have no solid evidence to suggest that *Nosema ceranae* is involved, but we do know that is in the US. Is it new? If you look back into the dark ages of honey bee disease research, you will find that Dr. Basil Furgala and I spent a good deal of time looking though microscopes at extracts of digestive tracts, looking for *Nosema apis* spores.

Once in a while, we would see something that looked like *N. apis*, but it was too small. Another difference was that the smaller organisms were sort of half transparent at one end and opaque at the other, not typical of *N. apis*. We thought maybe it was a different *Nosema* that might infect honey bee larvae or smaller spores that hadn't matured, but we just overlooked it and didn't count them in our studies.

A brief note in the Volume 45, Number 3 (2006) issue of the Journal of Apicultural Research has an article by I. Fries and cooperators showing light and electron microscope images of the spores of both species. They look similar to what we saw, but none pictured have the split color look.

Another potential factor in this disorder situation could be malnutrition. It is known that malnutrition is a serious stress on honey bees, leaving them more susceptible to diseases and complicating additional stresses, such as the physical beating taken while being hauled around on trucks, pesticide exposure, or food shortages while the bees are being used at saturation densities for crop pollination.

When foraging honey bees can not find and collect enough quality pollens to meet the nutritional needs of the colony, then the brood that is reared is done so at the expense of the nutrients that have been stored as reserves in the bodies of the nurse bees. The brood is not as healthy as it should be and neither are the nurse bees. Numbers of bees reared drops significantly when pollens are not available. The malnourished bees that are reared have shortened life expectancies. If this situation develops at the time when the winter bees are supposed to be reared, then it is likely that some of the colonies will lose all their bees as the summer

bees die of old age and the malnourished winter bees die early, also.

It has been the experience of some beekeepers that if you push malnourished bees to rear brood without incoming sources of fresh pollens, you can "burn the bees out" and the colony populations will dwindle soon thereafter. It appears that it would have been better to have not tried to stimulate early brood rearing.

The rapid disappearance of the bees seems remarkable. However, it must be remembered that during normal times, around 1,000 eggs are laid daily, 1,000 new bees emerge in a week, and 1,000 bees die six weeks later. Even with dead bee traps on the hives, only a few bodies are collected from the 1,000 that die each day. Nearly all the bees fly out and die away from the hive. So, if most of the bees are reaching the end of their life spans at about the same time, there can be a wholesale exodus from the hive.

Hopefully, the studies of various aspects of this disorder will provide us with a better understanding of how different factors influenced colony losses. Then, we can begin to consider how to prevent the losses in the future.

# **Bumble Bees in Almonds**

In the same issue of JAR, referred to in the previous article, researchers in Israel report on the effects of placing some bumble bee colonies in the corner of an almond orchard. The bumble bees stayed pretty close to home. Counts of bees in trees showed 0.7 of a bumble bee per tree right outside their nests, and 0.04 bumble bees in the trees at distances of 150 and 450 meters from their nests. Honey bees were visiting trees in the same areas at rates of 5.4, 9.1,

and 8.8 bees per tree. The fruit sets were 18.2, 15.9 and 14.0%, respectively. The authors concluded that the bumble bees were responsible for the improved fruit set near their nests, especially since the honey bee visitation was reduced in that area. [Editor's Note: Hand pollination of almonds can result in 40% set and good honey bee pollination will result in more than 20% set in California.] The authors concluded that adding more honey bees probably could have improved the set, but at least it appeared that bumble bees were contributing.

## 2006 CSBA Convention Review (Cont.)

### Oxalic Acid and Mite Control

At the well-attended Research Luncheon, Dr. Marion Ellis, from the University of Nebraska, reported on recent studies on the chemical and how registration was coming along.

First, information was provided on the effect of acid contact with mites and bees. It appears that the mites are 70 X more susceptible to oxalic acid than are the bees.

Second, Marion reviewed the different ways the acid is used to treat colonies. The most accepted way is "trickling" the formulated syrup into the spaces between the frames in autumn or early winter. If a sprayer is used, half the spray (or even all of it) should be applied from the bottom of the combs. It spreads around in the space better. The acid crystals can be placed in a device that quickly heats the crystals and drives the acid off as vapor. This appears to be best done when the colonies are pretty well "sealed up" for winter. Otherwise, the vapor escapes too fast to get the job done.

Both the spray (92%) and the trickle (87%) were quite effective. The cost (not

counting labor) is about \$0.04 - \$0.05 per treatment.

Next we were informed about the results of trying to eliminate *Varroa* from packages by spraying oxalic acid in sugar syrup. The bees were confined to the packages for 72 hours after treatment. Preliminary tests led to 63% control.

Then, to refine the application, over 100 cages of honey bees with mites were treated with varying volumes of oxalic acid per bee. Mite reduction was pretty good at 2.0 ml per 1,000 bees but exceeded 90% with higher volumes of syrup. However, at the 2.0 ml dose, a bit less than 20% of the bees were lost. Higher volumes of syrup pushed bee loss into the 30% range, which should be more bees than a beekeeper would be happy to lose.

Some actual field trials conducted in October led to somewhat spurious results. The numbers of mites per bee in the bulk container was determined. Those bees were used to make up packages. They were treated and installed. Just eight days later, the populations were re-sampled and the mites counted. The bulk bees stayed the same. The high and low dose oxalic bees had about an 80% mite loss. The control had a 22.5% increase.

Obviously, the mites could not have reproduced and increased their numbers by over 20% when there wasn't even any brood emerging in the colonies. So, robbing or colony collapses in the area must have been contributing mites in a big way.

There is an effort underway, with the American Beekeeping Federation as the applicant, to get EPA to allow treatments with oxalic acid as a "biopesticide." The EPA still hadn't responded (as of November 15<sup>th</sup>, 2006) to the pre-registration questions

coming from a meeting held back in February of 2006. The IR-4 (minor use registration program) is assisting in this effort.

Suggested packaging is: a 35 g packet (20 colonies), a 140 g packet (80 colonies), and a 700 g packet (400) colonies. A syringe and one-liter container could be sold with the packets or separately. Marion repeatedly mentioned wearing the proper protective equipment, even though the acid is not particularly dangerous if handled properly.

# **Honey Bee Breeding**

Soon to be UC Davis honey bee researcher (May, 2006) Sue Cobey explained to the group why it is important to maintain a large and varied genetic base for the health of the US beekeeping industry. Specifically, variation should reduce the susceptibility of our bees to diseases and parasites and should increase the ability of our bees to adapt to environmental changes.

Sue reviewed the mechanisms by which our current bees resist *Varroa*: 1. Varroa Reproductive Sensitive Hygiene (VRSH - used to be "SMR" bees – an additive trait), 2. "hygienic" bees, as from U Minn – a recessive trait – and 3. grooming behavior. Sue believes that since we lost most of our original feral colonies to tracheal and varroa mites, we have lost valuable genetic material that might have protected our bees. And, further, our bee breeders use only a relatively small number of breeder queens, even if you count all of them in the country. These are considered "genetic bottlenecks."

So, Sue would like to bring the semen of some European stocks into the US to invigorate her New World Carniolan line and perhaps other stocks at UCD. There is some German *A. m. carnica* stock with excellent

grooming behavior. There are some more Russian stocks that somehow reduce the growth rate of *Varroa* that feed on them. She currently is participating in stock improvement projects with Carniolan bees in New Zealand and Canada.

A portion of Sue's presentation was a defense against the criticism that instrument-tally inseminated (II) queens just can't compete with naturally mated ones, in terms of building populations, producing honey, and persisting. Five past studies have found II queens equivalent to naturally mated queens. Six have found II queens superior. But, one has demonstrated II queens to be inferior. Evidently, the last paper "got a lot of press."

Sue hopes to stay in touch with the geneticists who are teasing apart the honey bee genome. She hopes that they will be able to tell her which individuals to add to her breeding program, based on specific traits she wishes to add or avoid in her breeding. The genetic analysis now can be conducted on a wing tip, so you don't lose (destructive sampling) the queen or drone during the test.

Sue also hopes that we will learn how to preserve bee semen, frozen for long periods of time. Thus, Sue looks ahead to all the challenges and possibilities with true optimism. It will be great to have her affiliated with the UC Davis facility.

# 2-Heptanone, Revisited

Dr. Gloria DeGrandi-Hoffman returned to the podium to bring us up to date on the progress of getting 2-heptanone, a honey bee pheromone and beeswax solvent, to market as a varroa mite killer. Both CSBA and the Almond Board of California have provided funding for this project.

In cooperation with the USDA Bioproducts Research Lab in Albany, CA, they most recently have tried a wax and glucose covering. The mite kill is in the ballpark with Apistan strips. But, they still would like to find a better formulation. So far, no independent manufacturer or formulator wishes to produce the product.

In addition to killing mites, the 2-heptanone stops adult wax moths from laying eggs, stops robbing behavior between colonies, and is being tested to see if it inhibits egg laying by small hive beetles.

# Hygienic Behavior in Honey Bees

Dr. Marla Spivak was the third speaker who wished to impress the audience with the fact that honey bees bred for hygienic behavior are worth the money to purchase and will be important in disease and parasite control long into the future. However, most of the members of the colony population have to manifest the behavior in order for it to work well enough.

Hygienic bees have been determined to detect the odor of abnormal brood better than non-selected stocks. So, if we keep incorporating these genes into our bees, shouldn't we eventually have better bees? Marla thinks so, as do Tom Glenn and Sue Cobey.

Marla has begun to shift her focus in a different direction. Still under the umbrella of colony-level defense, she is taking a closer look at propolis and propolis collection. She and some other co-workers have demonstrated that propolis inhibits human HIV virus replication in two human tissue culture lines. One of Marla's students found Brazilian "green" propolis to be nearly as potent as tylosin against *Paenibacillus larvae* 

*larvae*, the microbe that causes American foulbrood disease.

Sample contents taken from honey bee crops of workers fed tylosin and green propolis demonstrated that the tylosin still was nearly 100% active, while the propolis already had lost about one third of its activity. Interestingly, the sugar syrup control showed no antibiotic activity when tested against the bacterium. After entering the crop, the syrup actually gained some antibiotic properties.

Thus, propolis seems to do the following for a colony: 1. seals cracks, 2. physically keeps out some intruders, 3. covers hive components with an antibiotic, and 4. may help the immune system of bees.

Although US beekeepers do not care much for propolis and select against its production, it still comes in. The main sources are trees of the *Populus* genus: poplars, cottonwoods, and aspen.

#### Soft Treatments for Varroa

Dr. Medhat Nasr, the provincial apiculturist from Alberta, Canada, shared with us information that he has gleaned from his own and others' studies on controlling *Varroa* with less toxic, and less contaminating chemicals, than we have been using in the past.

Medhat refers to our earlier chemicals as "Magic Bullets" or "Smart Chemicals." They could eliminate nearly all the mites in a colony, and not hurt the bees (too badly, we thought!!). We stuck in the strips and took them out, later (or not), and that was it. But, mite resistance to those acaricides put an end to that. Right now coumaphos resistance in Alberta varies from 18-75%. Going back to fluvalinate strips provides 40-85% control.

Acaricide residue in beeswax runs 2-3 ppm in Switzerland. It is similar for brood combs in Alberta colonies, and honey supers have about half as much residue in the wax. Previous analyses of wax from New Jersey combs showed coumaphos residues approaching 50 ppm and queens were failing badly.

So, we started using "Dumb Chemicals": organic acids, essential oils and sugar esters. Medhat showed about 10 different ways to apply formic acid. In a few cases the individual handling the acid appeared to wear gloves, but not all. Only a couple actually showed a respirator.

Of the various means of application, one thing stood out – the results were better in cooler temperatures (60-80% kill) than in warm (mostly 30-40%). That could very well relate to the amount of capped brood present, in which the mites would go unscathed.

Thymol is available in at least four formulations, in addition to straight crystals:

1. ApiLife VAR®, 2. Apiguard®, 3. Thymo Var® and Exomite®. Oxalic acid, formic acid and Apistan® all gave better results than ApiLife VAR in the data Medhat presented.

Seventeen days following treatments, the percentage of adult bees infested with mites stayed the same with Exomite, but increased with two different formic acid treatments and the control. The number of mites that fell to the sticky board was less than a thousand with Exomite, a bit over a thousand with the formic acid treatments, and less than a thousand in the control. An Apistan treatment killed about 4,000 mites in the Exomite colonies; 5,000 in the formic acid treated colonies; and 6,500 in the controls. Thus, all the treatments reduced the mites a bit, but there were many left in the colonies after treatment.

In his discussion of oxalic acid, Medhat showed more applicators in addition to those shown by Marion Ellis. In Canada, colonies wrapped for winter had better mite kill than those in the open with vaporized oxalic acid, but control varied from about 50% when the crystals were simply pushed in the entrance and heated vs. about 94% when the fumes were blown into the hive with a fan.

Given all this, Medhat had a suggested program for mite control. In spring if the average percentage of mite infestation is greater than 10% on adult bees from the brood nest, treat with oxalic acid or formic acid. In the summer, split the colonies and requeen with resistant stock. In fall, check for percent infestation of adult bees. If it exceeds 20%, then use oxalic acid vapor (should get both varroa and tracheal mites). If mite counts shoot up, resort to Apistan or CheckMite+.

# Hivastan® for Varroa Control

Doug Van Gundy, from Wellmark, International, shared information on a promising new varroacide for beehives. The active ingredient, fenpyroximate, is an acaricide currently being used in crop plant protection with a tolerance for food. The literature claims that the chemical is not toxic to honey bees by ingestion. (I hope they tested immature bees as well as adults!) The formulated product (0.3% a.i.) is a greaselike, milky colored material containing irradiated honey. It will be sold in three gallon buckets. The beekeeper will measure out eight ounces of the material, form it into the shape of a patty, and place it on wax paper or cardboard on the tops of the frames in the brood nest. A statement with the product claims that it melts at temps higher than 80 degrees. Maybe that is why the following complications occur when the

material hits 95 degrees in the brood nest area.

It appears to be a bit hard on the bees and brood during the first 24 hours that it is in the hive, causing some mortality. Then, things straighten out in the next 48-72 hours and things go back to normal. The treatment averages about 80% mite kill, but can vary from 60-100% probably depending upon the amount of brood in the colony.

Excerpts from ABRC or AHPA Presentations

### **USDA** Beltsville Disease Studies

Jeff Pettis shared some insights from their recent studies.

- 1. The researchers can detect nine different viruses in bees. Checking queens, they found 95% to have infections by multiple viruses and the other 5% had only one virus.
- 2. Inherent resistance to AFB in larvae depends upon whether or not their genes for resistance have been "turned on." Perhaps this can lead to determining what stresses the bees have encountered by seeing which genes have been turned on.
- 3. Probiotics, bacteria fed to animals to protect them from other infections, worked pretty well in the lab against AFB but, not in the field.
- 4. Results of a two year study that began with mineral oil on the top bars expanded to comparisons of mineral oil in towels, then other oils in towels. Eventually, Apiguard® proved to be better than bagged thymol oil, mineral oil, mineral oil plus thymol, and Exomite® (a specially formulated thymol dust), but each of the other treatments were better than the controls. Interestingly, in colonies that died, the larvae were succumbing to EFB. The causative microbe was nearly pure on culture plates,

which is not typical for honey bee larvae that die from EFB in untreated colonies.

Anita Collins brought us up to date on her advances in the area of preservation of honey bee semen. She said that maintaining 50% viability for 12 months would be "adequate" but less than desired. The sperm must be able to swim into, then out of, the spermatheca. Interestingly, fresh semen from drones varies from 5 to 95% viable, depending upon the drone. An average is about 73-80% alive for a single drone's semen in the collection syringe, but if you try to collect more semen into the same syringe, viability of all the sperm drops to 28-45%, probably from mechanical damage. So, it appears that washing the sperm off the drones into a sugar plus salt extender solution, allowing them to swim, then centrifuging them, will provide about 95% viability (from good drones).

#### Bee Schools

The Sacramento Area Beekeepers Association is sponsoring two beginning and one intermediate beekeeping classes in March and April. The cost is \$35 per adult, \$60 per couple, and \$15 per participant accompanied by an adult. All classes will be held at the Sacramento County Cooperative Extension auditorium at 4145 Branch Center Road (1 block west of intersection of Bradshaw with Keifer). Weather permitting, colonies will be examined, so bring at least a veil, if you have one.

Beginning Sessions will be held on Saturdays, March 24 and 31. Things begin at 8:00 AM and end at 5:00 PM, with an hour lunch break. Randy Oliver, bee breeder, pollinator, and bee journal author will teach these sessions.

The intermediate session will be held on Saturday, April 14<sup>th</sup>, with similar hours as

above. Course instructor will be Extension Apiculturist Eric Mussen from UC Davis.

For registration information, contact Sacramento Beekeeping Supply at (916) 451-2337 (Tues.-Sat.; 9:30-5:15) or e-mail sacbeek@csnet.com.

# Sincerely,

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