## FALL AND WINTER MANAGEMENT OF ALFALFA LEAFCUTTER BEES

#### Saskatchewan Alfalfa Seed Producers Association

By mid-August, alfalfa leafcutter bee females begin to die off and the population working in the field gradually decreases. Each female bee has produced many progeny, filling and capping several tunnels, which is evidenced by the filled nest blocks piled in the storage shed.

#### Alfalfa Leafcutter Bee Diapause

Most progeny of the domesticated alfalfa leafcutter bee species, *Megachile rotundata*, progress through egg and larval stages, spin their cocoons, and go into a prepupal state known as diapause. Diapause is a condition characterized by an increased concentration of glycerol (anti-freeze) in the body tissues which enables bee prepupae to survive low temperatures.

Many insects which go into diapause, including the alfalfa leafcutter bee, require a cold period to break diapause and allow for further development. Although diapausing bee prepupae will eventually continue development and emergence at 20 degrees C, the emergence period is from 98 to 342 days, or almost a year. The cold storage period results in a shortened incubation period and synchronized emergence, so that once incubation at 30 degrees C begins, the bees will generally emerge within an 18 to 25 day period.

### Second Generation in Alfalfa Leafcutter Bees

The alternative to diapause is continuous development into a second generation adult. The alfalfa leafcutter bee is normally a multivoltine species (capable of producing two or more generations per year). In some regions of the northwestern United States, two full generations and a partial third generation are produced. In Canada, one full generation is produced, often with a partial second generation depending on latitude, nesting material, and weather conditions. The determining factors for second generation development vs. diapause are unclear. Some researchers attribute the number of generations solely to temperature, while others have proven a genetic involvement. Certainly, there appears to be an higher level of second generation in a hot, dry summer than in a cooler year, but this may be a response of the adult female bee rather than the developing bee larva. It is likely that both environmental and hereditary factors play a role in second generation.

The net effect of any second generation in Canadian alfalfa leafcutter populations is probably to reduce the numbers of cells for storage. Whether second generation females have time to produce progeny and set seed will certainly depend on how soon they emerge and on the weather conditions when they do emerge. In central Washington, adult bees emerged within 38 days (1968) and 23 days (1970) from the time eggs were laid; the earliest adults emerged in mid to late July in the warmest seed-producing areas. This would seem to indicate that in most Canadian regions, with their shorter seasons and cooler temperatures, the second generation adults emerging from capped nesting boxes in storage prior to stripping, 50 to 70 days from the initial release, as well as pupae in various stages of development in their bee cell samples. Whether many bees have emerged previously and have been left in the field is hard to determine. The presence of males late in August is a good indication of second generation, since the first generation males die off during July.

It appears that second generation adults develop from the earliest eggs laid, and there seems to be little that can be done to control this phenomenon. Once the first nest blocks are brought into storage these emergent bees will continue their development as long as the storage temperature permits. After diapausing individuals have spun their cocoons and the storage temperature is decreased, second generation bees will stop developing and emerging; these bees will not survive the winter and will die during the storage period. Thus, cooling the earliest-filled nest blocks will halt emergence, but not development, of second generation adults.

Researchers and producers have observed that in general, all of the progeny in a tunnel will be either emergent second generation or diapause individuals, so it seems unlikely that many second generation individuals develop at the base of a tunnel and then chew through their diapausing siblings to escape. Therefore, the loss to the producer will probably be approximately equivalent to the number of emergent individuals. However, this loss is difficult to quantify. Samples taken after harvesting and tumbling the bee cells will no longer contain cells from which bees have emerged. Second generation estimates are therefore limited to counting those individuals actually present in the sample, without an estimate of those which emerged during the summer or fall.

## **Removing Nest Blocks From the Field**

Nest blocks with 70-80% capped tunnels are generally removed from shelters, mainly to protect them from weather, rodents, birds, and re-drilling into capped tunnels by female leafcutter bees. Depending on the date and on the number of bees still present, these filled nest blocks may be replaced with empty nest blocks. This is a management decision which becomes easier with experience. Too much nesting space will result in many partially completed tunnels, necessitating extra work in harvesting these nest blocks with little return. Too little nesting space, on the other hand, may result in loss of production. Most producers allow two or three tunnels per female and generally add nest blocks after the first filled nest blocks are removed from the field.

Once the alfalfa bloom is finished and the bee population noticeably decreasing, most of the nest blocks may be removed from the field. The nest blocks with fewest capped tunnels are left to the last. In a cool wet season, nest blocks should be brought in promptly since mould problems will develop rapidly; this is not as critical in hot dry weather conditions.

# **Storage Prior to Bee Cell Harvesting**

Approximately 20 degrees C is the minimum storage temperature required for development from the alfalfa leafcutter bee egg stage to the prepupal diapause stage. Nest blocks should be stored for three weeks at this temperature, or less time at a higher temperature, to allow all of the bee larvae to finish feeding and spin their cocoons. The bee cells will then be hard enough to withstand normal harvesting procedures. Cells should feel dry and hardened, not spongy, soft, or moist.

### **Mould Control**

To reduce or avoid mould problems it was once recommended that nest backing material be removed and nest blocks stood on their sides. This allows for air movement and evaporation of excess moisture from the backs of the nest blocks, which is especially necessary with polystyrene nest blocks in humid weather. Blocks are stacked in such a way as to allow air flow through the stack. This practice, while useful in controlling mould build-up, also creates a potential for rapid parasite build-up by making cells accessible to adults of the chalcid parasite, *Pteromalus venustus*, and other species which either come into the storage area in the nests or emerge during the pre-harvesting storage period. A compromise between mould and parasite control may be to maintain tight nests during the three week warm storage period, then remove the nest backs once temperatures are cooled so that any late-hatching parasites are inactivated by cool temperatures. Air circulation, exhausting of moist air, and the use of dehumidifiers may be necessary to lower moisture levels in nest blocks.

### **Drying Nest Blocks**

Polystyrene nest blocks may require active drying with air exhaust or dehumidifiers in order to facilitate harvesting of bee cells. The nest blocks should be kept at cool temperatures under conditions which promote drying until cells may be removed easily from tunnels throughout the nest block. The nest blocks easily take up moisture, and producers who have left the blocks in winter storage until May often find that they are once again very moist and difficult to harvest, with resulting crush damage to the bee cells. Producer experience suggests that the best time to harvest nest blocks is between November and March. The nest blocks must be stored until harvesting at normal winter storage temperatures of 5 to 8 degrees C both before and after bee cell removal, in order that the incubation period is not adversely affected.

#### **Fall Parasite Control**

Chalcid parasites found in alfalfa leafcutter bee nest blocks during the fall storage period are likely the second generation or emergent offspring of the field parasite population. These parasites can represent a threat to the leafcutter bee population since they will mate and attempt to parasitize developing bee larvae, causing a decrease in bee cell live count and an increase in numbers of parasitized bee cells. Unfortunately, the practice of removing nest backs to dry nests also allows easy access by parasites to potential hosts, by exposing the backs of the nest blocks and cracking the leaf seals around the capped tunnels. As discussed previously, this practice is not recommended until after the three week warm storage period is completed and temperatures have been cooled to below 15 degrees C.

Deployment of ultraviolet light / water-traps in the alfalfa leafcutter bee nest block storage area will help to control chalcid parasites in the fall. For ultraviolet light / water traps to be effective, the area should be insect-proof and light-proof. Emerging second generation adult bees will also fly to the ultraviolet light / water traps, as will moths and other insects. Studies on the use of dichlorvos resin strips for fall parasite control have indicated that exposure of nest blocks with backs removed to dichlorvos rates of 0.75 strip per 1000 cubic feet for up to seven days does not harm the alfalfa leafcutter bee prepupae within the nest blocks.

### Harvesting, Tumbling, and Storage of Bee Cells

Once all bee larvae have spun their cocoons, the storage temperature is reduced to below 15 degrees C to prevent further development and emergence of any second generation bees and parasites. Alfalfa leafcutter bee cells should be harvested, tumbled, and placed in cold storage as soon as possible to avoid problems with mould, parasites, stored product pests, and mice. Temperature fluctuations, which may affect the live count, must also be avoided.

There are many methods of harvesting cells from nesting laminates and nest blocks. The main points to watch for are that bee cells are not crushed or damaged and that the nesting laminates or nest blocks are not chipped or broken. Tumbling of harvested bee cells helps to remove empty cells and leaf debris, thus reducing volume. Tumbling also assists in the removal of pollen balls, mouldy cells, bee cadavers, and stored product pests. A major problem with many bee cell tumblers is the dust and mould spores which they release into the air. Producers may find themselves becoming more sensitive to this dust each year and in some cases serious allergies may develop. It is strongly recommended that tumbling be done outside or in an open shed with good ventilation, or that the tumbler be connected to a vacuum system which vents outside. Face masks or repirators should also be worn for added protection.

Commercial cell breakers are also available. The cell breaker breaks sequences of cells into single cells, on the premise that emerging leafcutter bees will be able to chew out of their individual cells without having to pass through a sequence of cells and possibly contact developing bees or diseased bee cadavers. The cell breaker is also useful in reducing bee cell volume by removing leaf debris and empty cells, and by crumbling pollen balls and bee cadavers. The cell breaker must be carefully adjusted to avoid damaging or crushing the bee cells. Bee cells which pass through a cell breaker should be decontaminated the following spring to destroy fungal spores which may be spread over cell surfaces during the cell-breaking process.

After harvesting and tumbling operations, alfalfa leafcutter bee cells should be placed in containers, closed up, and placed in cold storage. Large green garbage bags, plastic pails, cardboard drums, and cardboard boxes have all been used to successfully store bee cells. Containers should have an adequate seal so that the cells do not absorb moisture.

Cells should be stored at 5 to 8 degrees C to maintain diapause and render stored product pests inactive. Storage chambers range from refrigerators, garages, basements, and root cellars to specially-designed walk-in coolers. The walk-in cooler may be a modification of the incubation chamber, and will be best for temperature and humidity control. The storage facility depends on the size of the operation, what is available, and what is affordable.

Containers of alfalfa leafcutter bee cells must be stacked to allow air flow among the containers. A tight pile of containers may allow bee cells in the centre of the pile to begin heating, causing bee prepupae to break diapause and develop to the adult stage. Once diapause is broken due to accidental heating, re-cooling of the bee cells will cause mortality in many of the prepupae which have begun to develop. Stored alfalfa leafcutter bee cells should be checked regularly for problems with heating, moisture, and mould. If the bee cells become damp or mouldy, they may be spread out to dry and re-packed when they feel dry and hard again.

# Sampling Alfalfa Leafcutter Bee Cells for Quality

Sampling the alfalfa leafcutter bee population is the only way to maintain and increase the quality of the bees. Bee cell lots may be mixed together or kept separate according to the origin of the stock, type of nest material, individual shelter, or location of the field. A random sample of each bee cell lot should be kept for analysis after the cells have been tumbled and stored. Bee cell samples may be sent by alfalfa seed producers to the Canadian Cocoon Testing Centre (Brooks, Alberta) for independent analysis.

Bee cells may also be analysed by the producer. To do this, several bee cell samples are weighed out and analysed, with the results then used to calculate the number of bee cells per unit weight. Each bee cell is carefully cut open to expose the cell contents. Razor blades and utility knives work well for this purpose. The cells are separated into the various categories such as the following:

- live bee prepupae
- dead bee larvae / prepupae
- second generation bees
- parasitized bee cells
- pollen balls

The "live bee prepupae" totals are used to determine the number of healthy bee cells per unit weight. The live count is then calculated using the appropriate formula as outlined below, with examples given for sampling 30 grams of bee cells ( $2 \times 15$  gram samples) and for sampling 60 grams of bee cells ( $4 \times 15$  gram samples):

Number of live bee prepupae per 30 grams x 15.14	= Live count per pound
Number of live bee prepupae per 60 grams x 7.57	= Live count per pound

In addition to determination of live count, the alfalfa leafcutter bee sex ratio can be determined by incubating a random sample of bee cells. Small incubation trays can be constructed from 10 x 10 grids of egg crate light diffusion material, covered on each side with a piece of glass or plexiglass. Each tray will hold 100 cells. An incubator may be set up in an enclosed space where a constant temperature of about 30 degrees C can be maintained. Bee cell incubation tests should be done after December 01 to give alfalfa leafcutter bee prepupae an adequate cold storage period; otherwise, the incubation period will be prolonged.