



PARAFORMALDEHYDE FUMIGATION FOR SURFACE DECONTAMINATION OF ALFALFA LEAFCUTTING BEE CELLS

The Saskatchewan Alfalfa Seed Producers Association has obtained an amendment of the paraformaldehyde label to include fumigation of alfalfa leafcutting bee cells for control of moulds (including chalkbrood), yeasts, and bacteria on bee cell surfaces. Paraformaldehyde is a toxic substance and the fumigation method outlined in this publication should be followed carefully. Because problems may occur with persistence of formaldehyde vapor under certain conditions, treatment of bee cells should only be undertaken in a building set aside specifically for paraformaldehyde fumigation and not used for any other purpose.

INTRODUCTION

Analysis of alfalfa leafcutting bee cells from numerous populations has shown that the surfaces of these cells are inhabited by a wide range of mould, yeast, and bacterial species (microflora). This microflora may adhere to the body of the leafcutting bee upon emergence from the cell, and thus be transferred into nest material where it can cause spoilage of provisions and contribute to bee mortality. Some of the moulds found on bee cells (e.g. *Alternaria*, *Aspergillus*, *Penicillium*, and *Rhizopus* species) may also be hazardous to the health of alfalfa seed producers, particularly during bee incubation and harvesting operations. At these times, holding high numbers of exposed bee cells in confined areas may result in unsafe levels of airborne mould spores in the incubator or workroom. Incorporation of decontamination techniques is important in order to reduce microflora present on leafcutting bee cells.

EFFICACY OF PARAFORMALDEHYDE FUMIGATION FOR DECONTAMINATION OF LEAFCUTTING BEE CELLS

Research to develop new technology for controlling moulds, yeasts, and bacteria on the surface of alfalfa

leafcutting bee cells has led to the discovery of paraformaldehyde as a fumigant which is highly efficacious in decontaminating the cells.

Paraformaldehyde is a white, solid polymer of formaldehyde which gives off formaldehyde gas when heated. In fumigation tests on leafcutting bee cells, evaluation of control and fumigated cell samples indicated significant control of moulds (90.4% to 100%), yeasts (99.9% to 100%), and bacteria (94.3% to 100%) on the cell surfaces. These data demonstrate that fumigation with paraformaldehyde at a rate of 20 g/m³ will provide control of bee cell microflora. Moulds including *Aspergillus* and *Rhizopus* species, along with most yeast and bacterial species, were almost completely eliminated.

Incubation tests of control and paraformaldehyde fumigated bee cells yielded no significant differences in percent emergence (99.0% in both control and treated cell groups) or time to emergence of adult bees (control cells 23.6 days, range 20-29 days; treated cells 24.0 days, range 20-29 days). This indicates that fumigation of bee cells with paraformaldehyde does not adversely affect pupal development of the leafcutting bee.

FUMIGATION OF BEE CELLS SHOULD BE UNDERTAKEN IMMEDIATELY PRIOR TO BEE INCUBATION. The method developed for fumigating bee cells is outlined below. Note that extreme care should be taken in handling paraformaldehyde; adequate ventilation following its use is essential. **Under no circumstances should paraformaldehyde be exposed to an open flame.**

METHOD FOR FUMIGATION OF ALFALFA LEAFCUTTING BEE CELLS

1. Place bee cells in incubation trays in well-sealed fumigation chamber (**BUILDING USED ONLY FOR PARAFORMALDEHYDE FUMIGATION**) and condition room for 48 hours at 20-25°C with a relative humidity of 60-70%. Avoid spraying water around the chamber; condensation on walls and standing water on

the floor may cause paraformaldehyde gas to precipitate during the fumigation process. Stack trays in racks which will permit air movement between trays.

2. Fumigate bee cells with paraformaldehyde prills (91-97% formulation) at a rate of 20 grams of product per cubic metre of fumigation chamber area (1.1lb./1000ft³) by placing product in an electric frying pan attached to an electric timer. Paraformaldehyde prills should be handled with caution; the use of gloves, eye protection, and a dust mask or respirator is advised.

3. To begin the fumigation process, set frying pan to maximum heat setting, set timer to provide power for 4 hours, then seal and lock the chamber and post a warning sign. **UNDER NO CIRCUMSTANCES SHOULD THE CHAMBER BE RE-ENTERED AFTER ONSET OF FUMIGATION.**

4. From onset of fumigation, allow a 24 hour period for formaldehyde gas treatment. After this 24 hour period, begin continuous ventilation by exhausting from the top of the chamber for a 48-72 hour period. Ensure that there is an adequate incoming flow of fresh air during ventilation.

5. **OPEN AND RE-ENTER THE CHAMBER ONLY AFTER COMPLETION OF ADEQUATE VENTILATION.** Use a full-face NIOSH approved respirator with formaldehyde or acid gas cartridge and particulate filter; coveralls and gloves should also be worn.

6. Following adequate ventilation, transfer trays of fumigated bee cells to the incubator. **ENSURE CONTINUING VENTILATION DURING THE BEE INCUBATION PERIOD BY USING EITHER AN AIR-TO-AIR HEAT EXCHANGER OR ADEQUATE FRESH AIR VENTILATION. THIS IS IMPORTANT BECAUSE THE BEE CELLS AND INCUBATION TRAYS MAY ABSORB FORMALDEHYDE GAS DURING FUMIGATION AND RELEASE THIS GAS DURING INCUBATION.** Continue ventilation during the first 5 to 7 days of incubation at a level which will allow maintenance of incubation temperature without allowing a build-up of formaldehyde gas residue sufficient to cause irritation of eyes or nose. Adequate fresh air intake during ventilation is important, as is internal air circulation with fans.

NOTE: Paraformaldehyde prills, 91-97% (manufactured by Hoechst Celanese) are available in 50lb. bags; they should be stored apart from other combustibles in a cool, dry place with adequate ventilation. Before handling, the user should be familiar with safety information contained on the paraformaldehyde label and in the MSDS (material safety data sheet) for paraformaldehyde.

SUMMARY

Research on the use of paraformaldehyde fumigation for control of microflora on the surface of alfalfa leafcutting bee cells has indicated that treatment at a rate of 20 g/m³ is highly efficacious for decontamination of bee cells. Paraformaldehyde fumigation of cells prior to incubation is not deleterious to pupal development of the bees during the incubation period and bee emergence is not adversely affected. Adequate ventilation in the fumigation chamber and in the incubator is essential following fumigation of bee cells. Use of mould control techniques including paraformaldehyde fumigation will assist in reducing microflora in leafcutting bee populations and increase the viability of these populations.

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SASPA Extension Publication No. 93-01
October, 1993

This research was funded by the National Research Council (NRC/IRAP), the Saskatchewan Agriculture Development Fund (ADF), and the Saskatchewan Alfalfa Seed Producers Association (SASPA), and is also supported by Agriculture Canada.