Donahaye, E.J., Navarro, S. and Varnava, A. [Eds.] (1997) Proc. Int. Conf. Controlled Atmosphere and Fumigation in Stored Products, 21–26 April 1996, Printco Ltd., Nicosia, Cyprus, pp. 169–174

CONTROL OF BEESWAX MOTHS USING CARBON DIOXIDE IN FLEXIBLE PLASTIC AND METAL STRUCTURES

B. YAKOBSON[1], S. NAVARRO[2], E.J. DONAHAYE[2],
A. AZRIELI[2], Y. SLAVEZKY[3] AND H. EFRAT[3]
[1]*Kimron Veterinary Institute, Bet Dagan 50 250, P.O. Box 6, Israel*
[2]*Department of Stored Products, Agricultural Research Organization, The Volcani Center, Bet Dagan 50-250, P.O. Box 6, Israel*
[3]*Ministry of Agriculture Extension Service, Animal Husbandry Division on Beekeeping, Hakirya, Tel Aviv 61070, Israel*

ABSTRACT

The building structure of bee hives is based on the wax cells produced by the bees themselves. The most important of the species that attack beeswax is *Galleria mellonella* (L.), the greater wax moth, and protection of stored combs from this insect is of high priority, especially in hot climates.

A new approach to control of this pest has been to replace the traditionally used chemical insecticides, such as EDB, by environmentally sound methods. We propose the use of controlled atmosphere methodology with CO_2 for insect control and subsequent protection of honey supers during storage. The results of field tests using mixtures of CO_2 in air at initial concentrations ranging from 96 to 63%, for prolonged exposure periods, are presented. The tests were undertaken using 12-m^3 plastic enclosures and specially sealed 34-m^3 20-foot freight containers. Both methods enabled complete control of the wax moths and subsequent protection from reinfestation. The rigid structure of the containers had the inherent advantage of giving better protection from rodents and pilfering. However, each container requires specific sealing procedures followed by sealing tests. The plastic enclosures, found to be suitable for smaller enterprises, had the advantage of ease of portability and installation at the storage site. Their structural seals were checked during manufacture. So far they have been under continual use for 5 years.

INTRODUCTION

A number of moth species, the most important of which are the greater wax moth, *Galleria mellonella* (L.), and to a lesser extent the lesser wax moth, *Achrois grisella* (F.), attack beeswax. They cause major losses to commercial beekeepers every year by damaging wax combs in storage (Eckert, 1951; Smith, 1990; Singh, 1962). Wax moths can cause rapid and complete destruction of unprotected combs. Since the combs can only be made by the honeybees themselves, beekeeing ventures can become losing propositions without a ready supply of empty combs during the brood rearing and honey flow season.

In the past, the accepted method for controlling these pests was fumigation with ethylene dibromide or methyl bromide (MB) (Burges, 1978). Since 1983 the use of ethylene dibromide has been banned because of its carcinogenic properties, while MB, as a result of the unprecedented international cooperation for ridding the world of ozone-destroying chemicals, was listed at the Montreal Protocol (UNEP, 1992) among the chemicals that contribute to the depletion of the ozone layer of the atmosphere. For developed countries, complete phase-out of MB is programmed for the year 2010 (UNEP, 1995).

The use of modified atmospheres (MA's) as a fumigation alternative that retains the special capacity for *in-situ* treatment of agricultural commodities has been developed to control storage pests (Navarro, 1987; Navarro and Donahaye, 1990). MA technology has been considered for wax moth control; however, only scanty information has so far been published on the effect of exposure period and MA concentrations on wax moth mortality (Cantwell *et al.*, 1972; Greatti and D'Agaro, 1992).

On-farm storage of honey combs is problematic since storage structures are generally not sufficiently sealable for an effective gas composition of the treated storage space to be maintained in order to both obtain control of the pests and prevent subsequent reinfestation. Therefore, to implement the MA technology, appropriate storage structures need to be developed (Yakobson *et al.*, 1990). Such structures should be sufficiently gastight to maintain the desired MA concentrations and, at the same time, should be durable and resistant to harsh environments. Such structures have been developed for the preservation of grain and are in use (Donahaye and Navarro, 1989). However, similar structures are lacking in the beekeeping profession. This study reports on both the development and on-site testing of a flexible structure and the adaptation of old transport containers, taken out of circulation, as MA treatment and storage facilities for beehive supers and combs.

MATERIALS AND METHODS

The commercial-scale field trials to control wax moths using carbon dioxide (CO_2) were based on laboratory studies undertaken to examine the sensitivity of all stages of *G. mellonella* to 60, 70 and 80% CO_2 in air at 25 and 30°C. These studies are now being summarized and will be published elsewhere.

Plastic treatment enclosures

The structure. Of the several prototype fumigation enclosures investigated, the one reported on here consists of a flexible 0.83-mm liner, made of reinforced chlorinated polyethylene (CPE), welded to the shape shown in Fig. 1. The enclosure was provided with a walk-in flap-door, zip-closed with a tongue-and-groove gastight zipper, and an inner protective curtain of netting to prevent ingress of bees when the flap was open. The

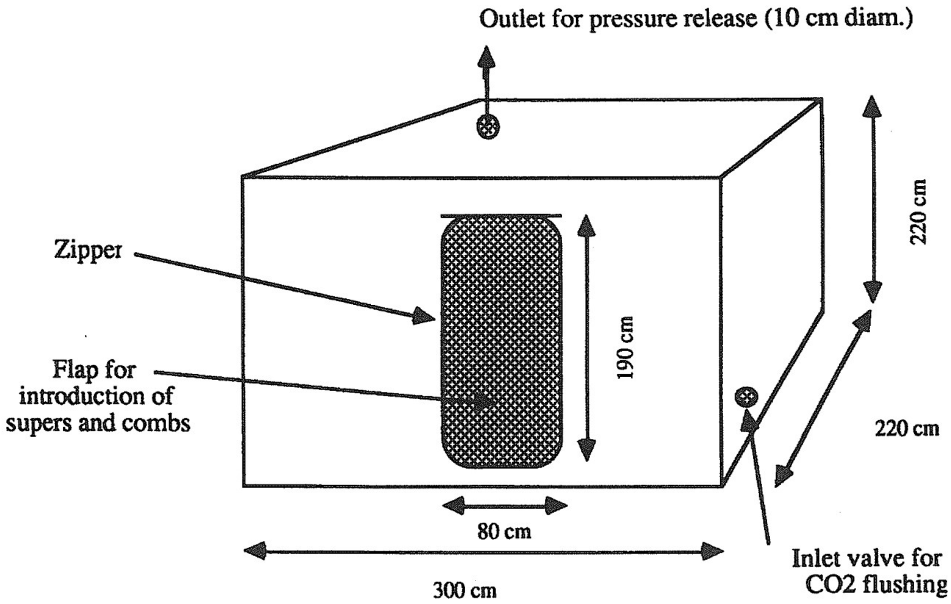

Fig. 1. Plastic enclosure for MA treatments of beehive supers (to control wax moths) and subsequent protection from infestation during storage.

liner was characterized by its ability to withstand prolonged periods of intense solar radiation. The enclosure was structurally supported by sets of vertically and horizontally positioned 19-mm fibre-glass rods held in place by connecting sockets in the corners of the liner. In this trial the enclosure was designed to contain 120 Langstroth-type supers holding wax combs. The supers were removed from bee hives in late spring, and after the honey had been extracted they were placed directly in the enclosure.

The treatment. CO_2 in the liquid phase was applied as a single shot at a rate of up to 6 kg/min from a pressurized cylinder through a release valve and pressure hose attached by snap-on connectors to the inlet valve of the enclosure. CO_2 concentrations were monitored during release (using a Gow-Mac thermal conductivity gas analyzer) until a concentration of approximately 65% CO_2 was obtained within the enclosure. This was equivalent to about 18 kg liquid CO_2 removed from the cylinder. Concentrations were measured daily for the first month. Then the gastight seal of the door was broken to permit the first inspection for infestation, and from then on the enclosure served as a physical barrier to prevent reinfestation.

Monitoring for infestation. Among the supers in the enclosure, one was heavily infested initially with all developmental stages of the wax moths. The wax combs in this super were examined monthly for 5 months for the presence of live adults, eggs, larvae and pupae.

Modified containers

This trial was carried out on one of several 20-foot freight containers that had been taken out of service and purchased by commercial beekeepers for storage of supers. The containers were then modified to form treatment chambers. Modifications consisted of installation of an inner PVC liner over the floor; sealing of cracks and crevices, particularly around the doors, with acrylic and silicon sealer; installation of a 4-mm diameter gas inlet duct, equipped with ball-valve tap and snap-on connecter, and an upper 10-cm diameter outlet duct sealed with a screw cap; installation of a pressure release valve; and installation through the container wall of two 4-mm diameter gas sampling lines, one leading to the top and the other to the floor. In the trial container reported on here, because the door gaskets were in poor condition, an additional inner door, consisting of PVC sheeting attached to the walls and held in place between sections of aluminium profile bolted around the opening of the container, was made. The beehive supers were progressively loaded into the container as they were removed from the hives in late spring for the extraction of honey. An initial examination, before the container was closed and sealed, revealed considerable wax moth activity on many of the combs.

Pressure tests. Following the above modifications, after the doors had been closed and sealed the container was subjected to a pressure test by creating a positive pressure of 50 mm water, using a portable vacuum cleaner, and recording the time taken for pressure to decay to 25 mm water.

Treatment. CO_2 in liquid form was flushed into the container from inverted cylinders. The outlet pipe was then capped and the container was kept sealed for 21 d. Then the doors were opened and a thorough inspection was made of supers and combs. Combs were removed from the container for additional laboratory examination.

RESULTS

Plastic treatment enclosure

Measurements of CO_2 concentration in the enclosure are given in Fig. 2. It can be seen that the CO_2 concentrations dropped from an initial level of 63% to 43% after 9 d. The first inspection of combs in the enclosure, made 30 d after fumigation, revealed 100% mortality of all wax moth stages in the infested supers. No further wax moth activity or damage was observed in the supers for 5 months following the treatment with CO_2.

Modified containers

Pressure test. The pressure decay test from 50 to 25 mm water revealed a halving time of 48 sec. Additional attempts to improve the seal around the door, followed by a second pressure decay test, resulted in an increase in halving time to 1.2 min.

CO_2 treatment. Three cylinders, each containing 26–28 kg CO_2, were emptied at flow rates ranging from 0.7 to 1.2 kg/min. After the third cylinder had been emptied, CO_2 readings were 91% and 96.5% at the top and bottom of the container, respectively. The

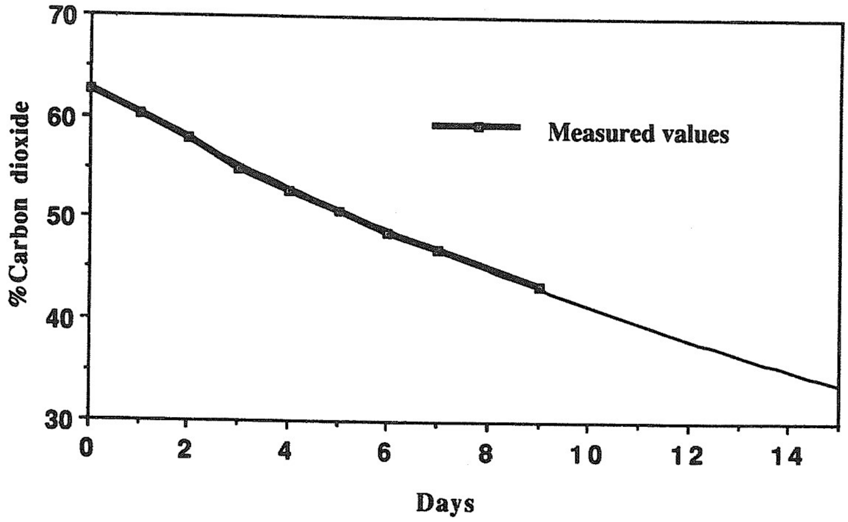

Fig. 2. Carbon dioxide concentrations recorded in plastic enclosure during the first 9 d of treatment.

CO_2 concentrations recorded over the first 10 d of the post-treatment period are shown in Fig. 3. This reveals that CO_2 concentrations decreased at about 5.4% per day.

At the inspection made upon opening the container after 21 d, no live insects were observed. Six combs that were taken from different regions of the container and held in the laboratory inside sealed plastic bags for a further 30 d revealed no live infestation upon examination.

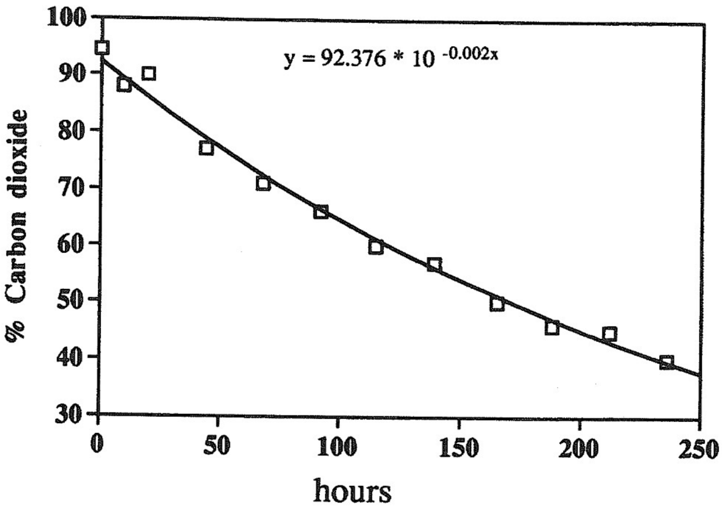

Fig. 3. Carbon dioxide concentrations recorded in modified freight container during the first 10 d of treatment.

CONCLUSION

One CO_2 treatment per season of the wax combs stored in a portable CPE fumigation chamber effectively controlled the wax moths. The modified freight container was also effective in controlling existing infestation and preventing wax-moth reinfestation and damage. Although the sensitivities of the developmental stages of the wax moths to different CO_2 concentrations under various climatic conditions have still to be investigated, it is clear from these field trials that very prolonged exposure to a range of high CO_2 concentrations was sufficient to obtain complete kill. This "single shot" CO_2 flushing treatment represents a user-safe and environmentally friendly alternative to fumigation with MB or phosphine.

ACKNOWLEDGEMENT

We would like to thank Yishai Ophir, apiculturalist of Moshav Hogla, for the cooperation and help extended to us during the course of this work. We are also indebted to Ben Ami Zilberstein of the Institute of Agricultural Engineering, ARO, for assistance in modification of the container.

REFERENCES

Burges, M.D. (1978) Control of wax moth: Physical, chemical and biological methods. *Bee World* **59** (4), 129–139.

Cantwell, G.E., Jay, E.G., Pearman, G.Jr. and Thompson, J.V. (1972) Control of the greater wax moth *Galleria mellonella* (L.) in comb honey with carbon dioxide. Parts I and II, *Am. Bee J.* **112** (9), 302–303; 342–344.

Donahaye, E. and Navarro, S. (1989) Volcani Cubes — a storage system for bagged or crated commodities. *GASGA Newsletter* No. 13, 3.

Eckert, J.E. (1951) Beekeeping in Hawaii. *Gleanings in Bee Culture* **79,** 393–400, 468–472, 509.

Greatti, M. and D'Agaro, M. (1992) Control of *Galleria mellonella* and *Achroia grisella* with carbon dioxide. *Apicoltore Moderno* **83** (4), 123–128 (in Italian).

Navarro, S. (1987) Application of modified atmospheres for controlling stored grain insects. In: *Proc. British Crop Protection Council, No. 37, Stored Products Pest Control.* Univ. of Reading, 229–236.

Navarro, S. and Donahaye, E. (1990) Generation and application of modified atmospheres and fumigants for the control of storage insects. In: *Proc. Int. Conf.: Fumigation and Controlled Atmosphere Storage of Grain* (Edited by Champ, B.R., Highley, E. and Banks, H.J.), Singapore, 14–18 February 1989, ACIAR Proc. No. 25, 152–165.

Singh, S. (1962) *Beekeping in India.* Indian Council of Agricultural Research, New Delhi.

Smith, F.G. (1960) *Beekeping in the Tropics.* Longmans, Green, London.

UNEP (1992) *Methyl Bromide — Its Atmospheric Science, Technology and Economics.* Montreal Protocol Assessment Supplement, June 1992, UNEP, Nairobi. 41 pp.

UNEP (1995) *UNEP 1994 Report of the Methyl Bromide Technical Options Committee (MBTOC) Assessment.* UNEP, Ozone Secretariat, Nairobi, Kenya. 304 pp.

Yakobson, B.A., Slavezky, Y., Navarro, S. and Donahaye, E. (1990) Application of carbon dioxide in portable fumigation chambers to control bee wax moths. In: *Proc. Int. Federation of Beekeepers Associations* (Edited by Ritter, W.), Gent, Belgium, 5–7 September 1990, 192–193.