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EFFECTS OF CARBON DIOXIDE AND COLD ANAESTHESIA ON THE HOARDING BEHAVIOUR OF THE HONEYBEE *

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Summary

Hoarding behaviour of groups of newly emerged adult honeybees was measured after they had been exposed to CO_2 for 10 min, or exposed to CO_2 for long enough to immobilize them, or exposed to cold. Both CO_2 and cold affected hoarding behaviour. Bees exposed to CO_2 for 10 min initially hoarded less, and later hoarded more, than controls. The hoarding rate of bees exposed to CO_2 only long enough to immovilize them, or exposed to cold, was generally depressed throughout the experiment. Due to the complex changes in hoarding that follow CO_2 treatment, cold is judged to be the preferred anaesthetic for use in hoarding experiments.

Introduction

Laboratory procedures for measuring nectar hoarding by the honeybee (*Apis mellifera*) have been devised (Free & Williams, 1972; Kulinčević & Rothenbuhler, 1973), and they have been used to study several factors influencing honeybee hoarding (Rinderer & Elliot, 1977; Rinderer & Baxter, 1978*a*; Rinderer & Baxter, 1978*b*; Rinderer & Sylvester, 1978; Sylvester, 1978; Rinderer & Baxter, 1979). The procedures involve measuring the rate at which a group of newly emerged adult caged honeybees remove sucrose solution from a gravity vial and store it in an empty piece of comb attached to the back of the cage.

Sometimes the bees are anaesthetized for transfer to the cages (Free & Williams, 1972; Rinderer & Baxter, 1979). Free and Williams found, in an experiment lasting 2 days, that CO_2 decreased hoarding whereas cold treatment increased it. Preliminary observations by Rinderer (unpublished), however, suggested that CO_2 had a different effect on the hoarding behaviour in the course of a 6-day experiment. Consequently, the effects, of CO_2 and of cold on the hoarding behaviour of newly emerged adult honeybees over a period of 6 days have now been investigated more fully.

Materials and Methods

General

In all three experiments in this investigation hoarding cages (Kulinčević et al., 1973) were each stocked with 30 adult worker bees (Rinderer & Baxter, 1978*b*) aged 0-24 h. All experimental treatments were administered on the same day that cages were stocked with bees. In all tests the amount of 50% (w/w) sucrose solution removed from gravity feeder vials in the cages was recorded daily for 7 days. The cages were held in a dark incubator at 35°C and 50% RH.

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Bees for the experiments were obtained from 8 different colonies. For any one replication of an experiment, 10 cages per treatment were taken from each of 3 colonies randomly chosen from the 8. Resulting data were submitted to analysis of variance and, where appropriate, least significant difference tests.

Effects of CO_2 anaesthesia

Five replications were made, in each of which 30 cages of bees were individually exposed to 100% CO_2 for 10 min in an enclosed chamber and another 30 cages were not so exposed.

Effects of different durations of CO_2 treatment

This experiment in two replications was designed to show whether different durations of exposure to 100% CO_2 resulted in similar effects. The same design as before was used, except that, additionally, 30 cages of bees were exposed to CO_2 only long enough (*c.* 2 min) to immobilize them (ETI).

Effects of cold anaesthesia

To assess the effect of cold anaesthesia on hoarding, bees were exposed to a temperature of − 20°C until they were immobilized (*c.* 3 min). Untreated cages of bees served as controls. Data were obtained from two replications of this experiment.

Results

Effects of CO_2 anaesthesia

Exposure to CO_2 for 10 min depressed hoarding in the first 3 days ($P < 0{\cdot}01$) and increased it in the next 3 ($P < 0{\cdot}05$) (Fig. 1). There was a weak interaction ($P < 0{\cdot}10$) between the factors of days and treatments, which reflects the different hoarding patterns through time of the treated and untreated bees. Over the whole experimental period

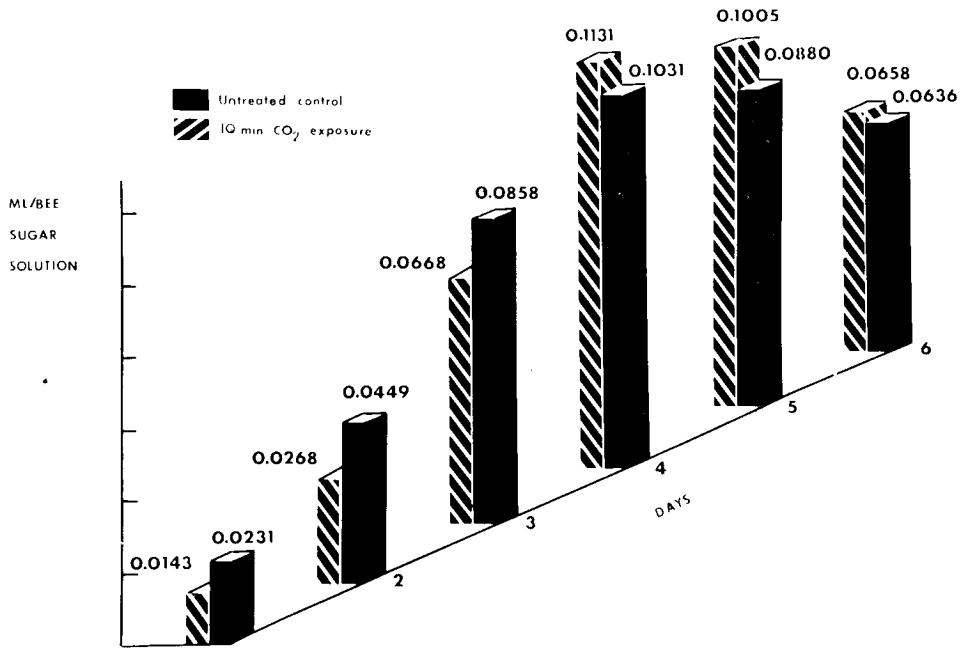

FIG. 1. Average daily hoarding rates (ml/bee) of caged bees, exposed to CO_2 for 10 min (hatched) and untreated (black).

there was little difference in hoarding between the two groups of bees, because of the offsetting effects of CO_2, which first depressed hoarding and later enhanced it.

There was also a highly significant ($P < 0{\cdot}01$) interaction between treatments and colonies, which showed that bees from the different colonies had different magnitudes of response to CO_2.

Effects of different durations of CO_2 applications

The hoarding patterns of bees receiving 10 min exposure to CO_2 and untreated bees were similar to those observed in the first experiment, but there was a different response by bees receiving only enough CO_2 treatment to immobilize them (ETI). The average total hoarding of the ETI bees was significantly ($P < 0{\cdot}05$) lower than that of bees with 10-min exposure to CO_2 (Fig. 2), although there was no significant difference in total hoarding between control bees and either type of CO_2-treated bees. Unlike the hoarding pattern of groups of bees receiving 10-min exposure to CO_2, the daily hoarding rate of ETI groups of bees was numerically less than the daily hoarding rates of groups of untreated control bees throughout the experimental period, except on 1 day.

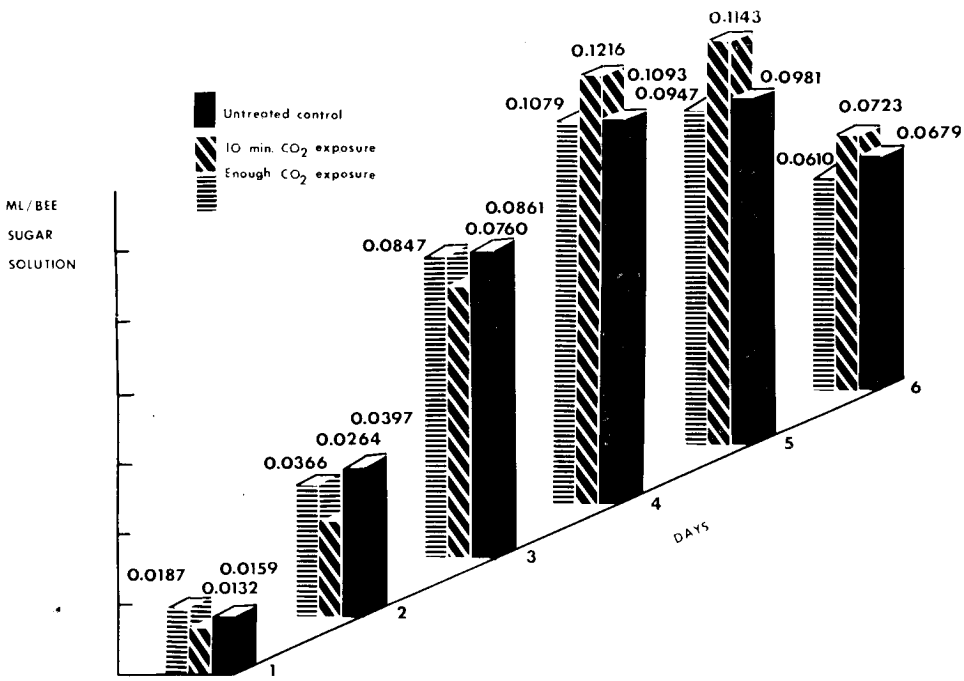

FIG. 2. Average daily hoarding rates (ml/bee) of caged bees, exposed to CO_2 for 10 min (hatched) for long enough to immobilize them (horizontal lines), and not at all (black).

Effects of cold anaesthesia

The effects of cold anaesthesia were more severe than those observed with CO_2 treatment; here was overall reduction ($P < 0{\cdot}05$) in hoarding (Fig. 3).

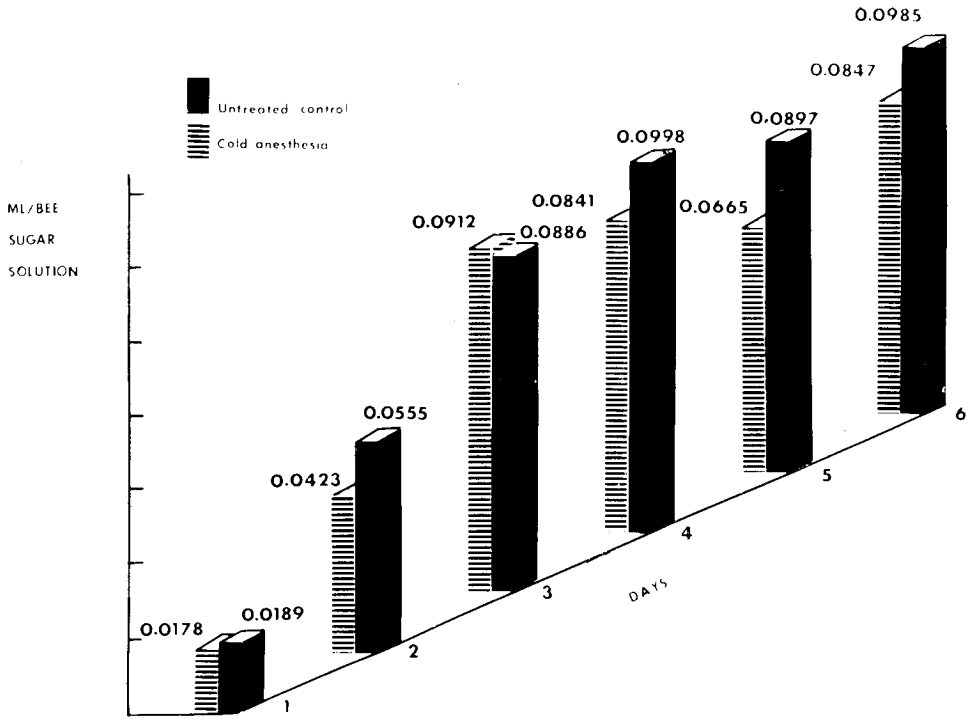

FIG. 3. Average daily hoarding rates (ml/bee) of caged bees, anaesthetized with cold (horizontal lines) and not aneasthetized (black).

Discussion

Taken collectively, these experiments suggest that after prolonged exposure to CO_2 there are two sequential effects. At first a toxic effect reduces hoarding; later, physiological changes occur that override the toxic effect and result in more rapid hoarding. A lesser exposure to CO_2 (ETI) apparently produces the toxic effect but not the later changes that accelerate hoarding. This hypothesis reconciles our CO_2 results with those of Free and Williams (1972).

The results with cold are more difficult to explain. Free and Williams (1972) found that cold increased hoarding, whereas we found that it decreased it. However, they used $-5°$ for 10 min and we used $-20°$ for *c.* 3 min, so experimental differences between the studies may be the source of the difference in results. An exploration of the causes of these differences could reveal the physiological underpinnings of hoarding behaviour.

The effects of cold on hoarding seem to be simpler and more consistent than those of CO_2, so cold is judged the anaesthetic of choice in experiments involving hoarding. Similar conclusions were drawn from experiments studying effects of CO_2 and cold on length of life (Tustain & Faulke, 1979) and foraging behaviour (Ebadi et al., 1980).

Acknowledgement

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