

## CONTROLLING RUSTY GRAIN BEETLES WITH CONTROLLED ATMOSPHERES

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### ABSTRACT

Mortality of adults, pupae, larvae, and eggs of the rusty grain beetle, *Cryptolestes ferrugineus* (Stephens), was determined at carbon dioxide (CO<sub>2</sub>) concentrations of 30, 40, and 60%, at a temperature of 30°C. The oxygen (O<sub>2</sub>) concentration was maintained constant at 10% and the relative humidity of gas mixtures was maintained at 75%. The gases were mixed from cylinders of CO<sub>2</sub>, O<sub>2</sub>, and nitrogen (N<sub>2</sub>) at a gage pressure of 35 kPa and the flow rate in an exposure unit was maintained at 55 mL/min. For a 60% CO<sub>2</sub> concentration in the atmosphere, 100% mortality of all the life stages of beetles was obtained in 3 days. For 30 and 40% CO<sub>2</sub> concentration in the atmosphere, all adults were killed within 8 days and all pupae, larvae, and eggs were killed within 4 days.

### INTRODUCTION

Controlled atmospheres can be used to control stored-product pests as an alternative to insecticides and fumigants. Pesticides excluding methyl bromide, phosphine, and malathion have been banned for use in stored-products by regulatory agencies in Canada because of their perceived adverse effects on human health and the environment (Bond, 1987). Many stored-product insects have developed resistance to a range of pesticides (Conway, 1987). In controlled atmosphere storage of grains the proportion of the normal atmospheric gases carbon dioxide (CO<sub>2</sub>), oxygen (O<sub>2</sub>), and nitrogen (N<sub>2</sub>), is altered to give a lethal atmosphere to stored-grain pests (Bailey and Banks, 1980).

The mortality of stored-product insects depends mainly on the type and concentration of gases in the mixture, the species and life stage of insects, exposure time, temperature and relative humidity (r.h.), (Bailey and Banks, 1980). Many researchers have conducted experiments to determine the mortality of specific stored-product insects in controlled atmospheres

(Lindgren and Vincent, 1970; Aliniaze, 1971; Navarro and Calderon, 1974; Calderon and Navarro, 1979; Spratt *et al.*, 1985; Soderstrom *et al.*, 1986).

Information on the mortality of rusty grain beetles to controlled atmospheres is needed to make this technique reliable and economical for application in Canada where rusty grain beetles are of great importance. Mortality of adult rusty grain beetles in elevated CO<sub>2</sub> concentrations (>60%) at temperatures common in Canadian grain was reported by White *et al.* (1988). Krishnamurthy *et al.* (1986) studied the mortality of adult rusty grain beetles for low CO<sub>2</sub> concentrations (<30%) and low O<sub>2</sub> concentrations (<2.6%) at 20°C. Rameshbabu *et al.*, (1991) studied the mortality of rusty grain beetle adults and eggs for elevated CO<sub>2</sub> (>60%) and depleted O<sub>2</sub> (<8%) concentrations at 10, 15, and 20°C.

The objective of this study was to determine the mortality of the adults, pupae, larvae, and eggs of the rusty grain beetle in CO<sub>2</sub> concentrations of 30, 40, and 60% at 30°C, 10% O<sub>2</sub>, and 75% r.h. The balance of the gas mixture was N<sub>2</sub>.

## **MATERIALS AND METHODS**

### **Experimental Apparatus**

A controlled atmosphere unit, developed by Rameshbabu *et al.* (1991), was used in the experiments with a few modifications (Fig. 1). Only one r.h. unit was used to humidify the gas mixture and eight exposure units were used at any one time. A rotameter (Gilmont Instrument Inc.) having a 0-305 mL/min range was used to measure the flow rate of O<sub>2</sub>, and two rotameters (Gilmont Instrument Inc.) of 0-2,200 mL/min range were used to measure the flow rates of CO<sub>2</sub> and N<sub>2</sub>.

### **Experimental Design**

Mortality of *C. ferrugineus* adults, pupae, larvae, and eggs was determined for CO<sub>2</sub> concentrations of 30, 40, and 60% at 30°C. The O<sub>2</sub> concentration and r.h. were maintained at 10% and 75%, respectively. The gases were released from the cylinders at a gage pressure of 35 kPa and the flow rate in the exposure units was maintained at 55 mL/min. Four replicates were run under each condition. Two replicates were used in the controls.

### **Experimental Procedure for Adults**

Two-month-old adults of *C. ferrugineus* were taken from laboratory cultures on wheat germ and whole wheat (1:19 by weight) at 30°C and 70% r.h. Twenty-five adults and 1 g of wheat germ were placed in each of the four insect boxes of an exposure unit and the controls. Insect boxes were assembled into exposure units and their inlets and outlets were connected to the gas lines from the r.h. unit and exhaust assembly, respectively. Flow meters were adjusted to set the proportion of individual gases (CO<sub>2</sub>, O<sub>2</sub>, and

$N_2$ ) for the gas mixture required in a given experiment. The composition of the gas mixture was analyzed at 12 hr intervals by injecting 5 mL samples into a gas chromatograph with a thermal conductivity detector and a fixed sample-loop volume of 1 mL (Hewlett-Packard model HP9850A, Avondale, PA USA). Relative humidity of the gas mixture was measured at 12 hr intervals using a Vaisala humicap, HM 1698 probe equipped with a thin polymer capacitance sensor and HMI 31 digital meter (Helsinki, Finland).

After each exposure period, one exposure unit was removed and its component insect boxes were dismantled. The flow meters were adjusted to maintain the flow rate at 55 mL/min in the remaining exposure units. The insects from the samples after treatment and those from controls were placed in 1 g fresh wheat germ in glass vials with perforated lids at 30°C and 70% r.h. and the number of live adults was recorded after 168 hr.

### Experimental Procedure for Pupae

Gelatine capsules containing 0.5 g ground wheat germ and last instar larvae of *C. ferrugineus* were maintained at 30°C and 70% r.h. until the larvae turned to pupae. Five pupae were put into each insect box of the exposure units and the controls. After every exposure the pupae were transferred to another capsule containing wheat germ and kept at 30°C and 70% r.h. until the emergence of adults. The emergent adults were counted.

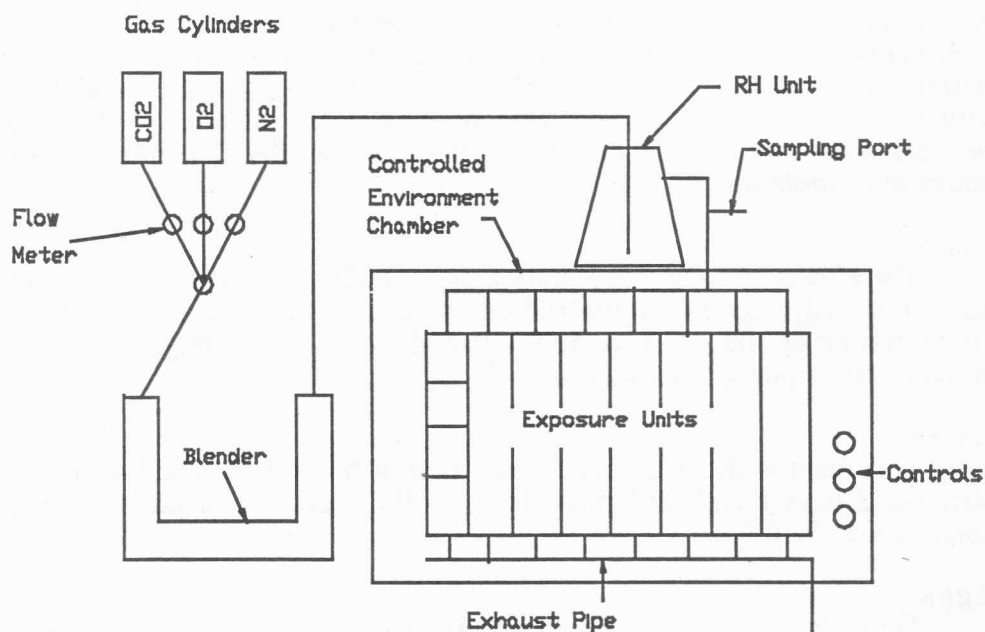


Fig. 1. Schematic diagram of the controlled atmosphere unit.

### **Experimental Procedure for Larvae**

Eggs of *C. ferrugineus* were collected from 250 adults placed on wheat flour and wheat germ maintained at 30°C and 70% r.h. The adults were sifted out and the culture maintained under identical conditions until the development of last instar larvae. Five larvae, each in a gelatine capsule containing 0.5 g wheat germ, were put into each insect box and the controls. After every exposure, the larvae were transferred to another capsule containing fresh wheat germ and kept at 30°C and 70% r.h. until the emergence of adults. The emergent adults were counted.

### **Experimental Procedure for Eggs**

Eggs of *C. ferrugineus* were collected from 250 adults placed on wheat flour maintained at 30°C and 70% r.h. Five eggs, each in a gelatine capsule containing 0.5 g wheat germ, were placed in an insect box and the controls. After every exposure, the eggs were transferred to another capsule containing fresh wheat germ and kept at 30°C and 70% r.h. until the emergence of larvae. The emergent larvae were counted.

## **RESULTS**

### **Adults**

The variations in CO<sub>2</sub> and O<sub>2</sub> concentrations were within ±2% and in relative humidity were within ±3% of the desired values. Mortality of adults was influenced significantly by the exposure period, CO<sub>2</sub> concentration, and their interaction. Treatment with a CO<sub>2</sub> concentration of 60% killed all adults within 3 days (Fig. 2). Treatment with CO<sub>2</sub> concentrations of 30 and 40% killed all adults within 8 days with no significant difference ( $P>0.05$ ) between these two treatments in terms of adult mortality. The mortality of adults in controls was 3-11%.

### **Pupae**

The effects of CO<sub>2</sub> concentration on mortality of pupae (Fig. 3) were not statistically significant ( $P>0.05$ ). Four days of treatment with a CO<sub>2</sub> concentration of 30%, 40%, or 60% killed all pupae completely. There was no mortality of pupae in the controls.

### **Larvae**

Treatment with a CO<sub>2</sub> concentration of 30% for 3 days and 40% and 60% for 2 days killed all larvae (Fig. 4). The mortality of larvae in the controls was 5-10%.

### **Eggs**

Three days of treatment with a CO<sub>2</sub> concentration of 30, 40, or 60% killed all eggs (Fig. 4). The effect of concentration of CO<sub>2</sub> on the mortality of eggs was not statistically significant ( $P>0.05$ ). There was no significant

difference among the treatments of 30%, 40%, and 60% CO<sub>2</sub> in terms of the mortality of all the life stages of the insect on the first day of exposure. The effects of exposure for 3 and 4 days on the mortality of all life stages for all the CO<sub>2</sub> concentrations studied did not differ significantly.

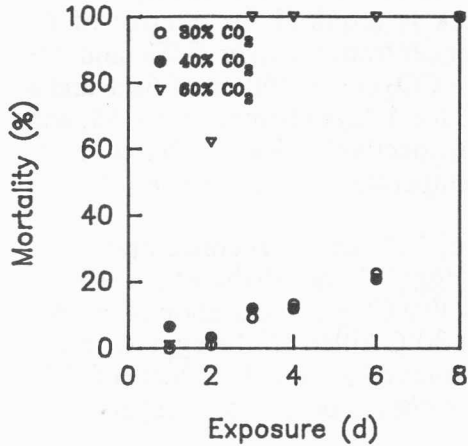


Fig. 2 Mortality of adults exposed to controlled atmospheres (10% O<sub>2</sub>; 75% rh; 30° C).

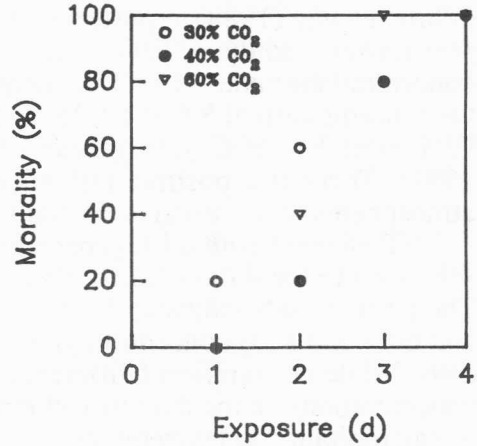


Fig. 3 Mortality of pupae exposed to controlled atmospheres (10% O<sub>2</sub>; 75% rh; 30° C).

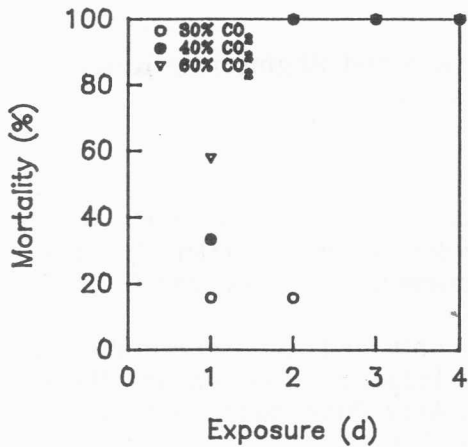


Fig. 4 Mortality of larvae exposed to controlled atmospheres (10% O<sub>2</sub>; 75% rh; 30° C).

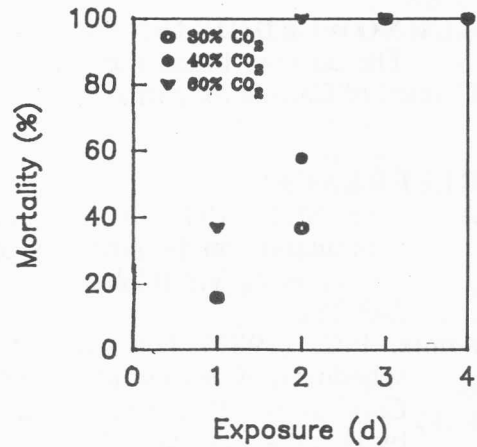


Fig. 5 Mortality of eggs exposed to controlled atmospheres (10% O<sub>2</sub>; 75% rh; 30° C).

## DISCUSSION

About 3 days are required for control of *C. ferrugineus* at 30°C under an atmosphere containing CO<sub>2</sub>:O<sub>2</sub>:N<sub>2</sub> (60:10:30%) at 75% r.h. Current moisture content (m.c.) recommendations for CO<sub>2</sub> treatment of grains at or below 14% m.c. and at or above 27°C, excluding those containing *Trogoderma* spp., are a CO<sub>2</sub> concentration of 60% for 5-6 days (Jay, 1984). White *et al.*, (1988) reported that 1 week is required for control of *C. ferrugineus* adults at 20°C with CO<sub>2</sub> concentrations over 54% and O<sub>2</sub> concentrations under 11%. Treatment with a CO<sub>2</sub> concentration of 68% and a O<sub>2</sub> concentration of 8% at 10, 15, and 20°C for 4 days resulted in 54, 58, and 73% mortality of *C. ferrugineus* adults, respectively (Rameshbabu *et al.*, 1991). There is a positive influence of temperature on the action of CO<sub>2</sub> atmospheres on *C. ferrugineus* adults.

Treatment with a CO<sub>2</sub> concentration of 70% and a O<sub>2</sub> concentration of 4% at 20°C for 4 days killed 50% of the eggs (Rameshbabu *et al.*, 1991). The present study indicates that treatment with a CO<sub>2</sub> concentration of 30, 40, and 60% and a O<sub>2</sub> concentration of 10% at 30°C killed all the eggs within 3 days. While no significant difference was observed among the effects of CO<sub>2</sub> concentrations on the mortality of eggs there does appear to be a dependence of egg mortality on temperature.

About 3-4 days are required for control of pupae and larvae, and 8 days for adults with a 30 or 40% CO<sub>2</sub> concentration at 30°C.

At low CO<sub>2</sub> concentrations (40%), adults of *Sitophilus oryzae* and *Trogoderma granarium* are the most susceptible to CO<sub>2</sub> among the life stages studied (Annis, 1987). In our study, we found adults of *Cryptolestes ferrugineus* to be the most tolerant of low CO<sub>2</sub> atmospheres.

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## REFERENCES

- Aliniabee, M.T. (1971) The effect of carbon dioxide gas alone or in combination on the mortality of *Tribolium castaneum* (Herbst) and *T. confusum* du Val (Coleoptera, Tenebrionidae). *J. stored Prod. Res.* 7, 243-252.
- Annis, P.C. (1987) Towards rational controlled atmosphere dosage schedules; A review of current knowledge. In: *Proc. 4th Int. Work. Conf. Stored-Product Protection*, Tel Aviv, Israel, Sept. 1986. (Edited by Donahaye, E. and Navarro, S.), pp. 128-148.

- Bailey, S.W. and Banks, H.J. (1980) A review of recent studies of the effects of controlled atmospheres on stored product pests. In: *Atmosphere Storage of Grains*, (Edited by Shejbal, J.), pp. 101-118. Elsevier Sci. Publ. Co., Amsterdam, Holland.
- Bond, E.J. (1987) Investigations on adverse properties of the fumigants methyl bromide and phosphine. 1. Residues of methyl bromide in fumigated commodities. In: *Proc. 4th Int. Work. Conf. Stored-Product Protection*, Tel Aviv, Israel, Sept. 1986. (Edited by Donahaye, E. and Navarro, S.), pp. 327-334.
- Calderon, M. and Navarro, S.. (1979) Increased toxicity of low oxygen atmospheres supplemented with carbon dioxide on *Tribolium castaneum* adults. *Entomol. Exp. Appl.* **25**, 39-44.
- Conway, J.A. (1987) A global view of arthropod pest status in relation to changing storage and marketing practice. In: *Proc. 4th Int. Work. Conf. Stored-Product Protection*, Tel Aviv, Israel, Sept. 1986. (Edited by Donahaye, E. and Navarro, S.), pp. 386-394 .
- Jay, E. (1984) Imperfections in our current knowledge of insect biology as related to their response to controlled atmospheres. In: *Controlled Atmosphere and Fumigation in grain Storages*, (Edited by Ripp, B.E. et al.), pp. 493-508. Elsevier Sci. Publ. Co., Amsterdam, Holland.
- Krishnamurthy, T.S., Spratt, E.C. and Bell, C.H. (1986) The toxicity of carbon dioxide to adult beetles in low oxygen atmospheres. *J. stored Prod. Res.* **22**, 145-151.
- Lindgren, D.L. and Vincent, L.E. (1970) Effect of atmospheric gases alone or in combination on the mortality of granary and rice weevils. *J. Econ. Entomol.* **63**, 1926-1929.
- Navarro, S. and Calderon, M. (1974) Exposure of *Ephestia cautella* (Walker) pupae to carbon dioxide concentrations at different relative humidities: the effect on adult emergence and loss in weight. *J. stored Prod. Res.* **10**, 237-241.
- Rameshbabu, M., Jayas, D.S. and White, N.D.G. (1991) Mortality of *Cryptolestes ferrugineus* (Stephens) adults and eggs in elevated carbon dioxide and depleted oxygen atmospheres. *J. stored Prod. Res.* **27**, 163-170.
- Soderstrom, E.L., Mackey, B.E. and Brandl, D.G. (1986) Interactive effects of low oxygen atmospheres, relative humidity and temperature on mortality of two stored product moths. *J. Econ. Entomol.* **79**, 1303-1306.
- Spratt, E.C., Dignam, G. and Banks, H.J. (1985) The effect of high carbon dioxide in air on *Trogoderma granarium*. *J. stored Prod. Res.* **21**, 41-46.

White, N.D.G., Jayas, D.S. and Sinha, R.N. (1988) Interaction of carbon dioxide and oxygen levels and temperature on adult survival and multiplication of *Cryptolestes ferrugineus* in stored wheat. *Phytoprotection*. **69**, 31-39.