

THE EFFECTS OF SUBLETHAL CONCENTRATIONS  
OF CARBON DIOXIDE (CO<sub>2</sub>) IN ATMOSPHERE ON  
SOME BIOLOGICAL ASPECTS OF THE COWPEA  
WEEVIL *Callosobruchus maculatus*  
F. (Bruchidae : Coleoptera)

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**Abstract**

Three sublethal concentrations of carbon dioxide in atmosphere (LC<sub>25</sub>, LC<sub>40</sub> and LC<sub>50</sub>) were investigated to study their effect on some biological aspects of *Callosobruchus maculatus*. At all sublethal concentrations the longevity of females was shorter than that of males. As the sublethal concentrations increased, the longevity of the survivor adults decreased. The number of eggs laid per female was greatly reduced as the concentration increased. These dosages did not inhibit eggs deposition. The reproductive potential of *C. maculatus* as measured by the mean number of offspring per female was considerably reduced by increasing the sublethal dose of CO<sub>2</sub> to LC<sub>50</sub>. These results were concluded from a progeny resulting from eggs exposed to the above mentioned concentrations for 3 days.

**INTRODUCTION**

In practice, the effectiveness of any control measure, chemical or non-chemical, is based on insect mortality, thus ignoring other effects that could be greater than

those shown by mortality data alone. Alterations in the biology of insects surviving from different chemical treatments had been indicated by several investigators (Parkin 1964; Reynolds *et al.*, 1967; El-Nahal and El-Halfawy 1973; Hashem 1981). Little work had been done on the effects of sublethal CO<sub>2</sub> concentrations in atmosphere on stored insects (Storey 1975, 1977, 1978; Spratt 1979a,b).

The present work is a study of the effect of sub-lethal doses of CO<sub>2</sub> in atmosphere on certain biological aspects of *Callosobruchus maculatus* F.

## MATERIALS AND METHODS

Females emerging from eggs treated with LC<sub>25</sub>, LC<sub>40</sub> and LC<sub>50</sub> of CO<sub>2</sub> in atmosphere at 30 ± 1°C and 70 ± 5 % R.H. were investigated from the view point of fecundity, longevity and fertility.

The parental insects were obtained from a laboratory strain maintained at the Stored-Product Insects Research Centre, Ministry of Agriculture. The stock culture was established by introducing 200 - 400 adults in a two pounds glass jar, half filled with broad beans and covered with muslin, held in place by rubber bands. The adults were permitted to oviposit in these cultures throughout their life span which did not exceed 12 days, and then removed, leaving the eggs glued to the surface of the beans. The emerging adults (about 500) were collected and released in two pounds glass jar, half filled with faba bean seeds. The adults were left to deposit eggs for 2 days, after which they were removed. Samples of bean seeds, that held 2-days old eggs were chosen from these cultures and were used in exposure tests for sublethal CO<sub>2</sub> in atmosphere. The experimental unit was 50 eggs glued to the surface of 10-15 seeds. This unit was prepared in a glass tube, 7 cm long and 1.5 cm diameter, covered in both ends with gauze. These tubes were introduced into a gas-washing bottle (dressel flask).

Three sublethal concentrations of CO<sub>2</sub> in atmospheres were used for treatment of 1-2-days old eggs of *C. maculatus*, these were :

LC<sub>25</sub> : 15.8 % O<sub>2</sub> , 63.2 % N<sub>2</sub> and 21 % CO<sub>2</sub>.

LC<sub>40</sub> : 15 % O<sub>2</sub> , 60 % N<sub>2</sub> and 25 % CO<sub>2</sub>.

LC<sub>50</sub> : 14.4 % O<sub>2</sub> , 57.6 % N<sub>2</sub> and 28 % CO<sub>2</sub>.

Bean seeds with eggs in gastight connected dressel flasks were held in a constant temperature room at 25°C and were exposed to the gas mixtures by leading gas out of the gas cylinder through copper tubes and a humidifying unit at 70% R.H. consisting of saturated NaCl/H<sub>2</sub>O solution into the flasks.

At the outlet of the flasks, O<sub>2</sub> content was determined continuously by Oxygen analyzer. After about 15 minutes (time for about 10 replacements of total flask volume by gas mixture) outlet concentration was identical with inlet concentration. The gastight sealed flasks were then separated from the gas mixture cylinder to permit further experiments with other flasks.

After 3 days from exposure, the flasks were aerated and the seeds were transferred to Petri-dishes in a culture room and examined for emerged adults at 25°C. Five pairs of the newly emerged adults from each treatment were prepared. The females of *C. maculatus* were easily separated from males by the last abdominal segment according to Shomar (1963), being emarginate with pygidium strongly convex at the sides with uniform pubescence in the male and not emarginated with median line of the pygidium supplied with white pubescence in the female. Each pair was placed in a muslin covered plastic vial (5 cm long and 2.5 cm in diameter) together with five seeds of uniform shape and size to serve as an oviposition site. These five seeds were changed daily by new ones so long as the female was alive and the removed seeds were incubated until the emergence of adults.

Through this technique it was possible to determine the total number of eggs laid and the total number of adults emerged per female. Each treatment had its own control. The daily changing of the 5 seeds of each pair was carried out as quickly as possible so as to minimize the time of exposure of the adults to laboratory conditions. This was done by quickly transferring the pair of weevils of each vial to a clean one containing new seeds. This technique was nearly similar to that of Gokhale and Srivastava (1975) in their studies on the ovipositional behaviour of *C. maculatus*.

## RESULTS AND DISCUSSION

### Effect on fecundity

As illustrated in Fig1, the effect of various sublethal concentrations of CO<sub>2</sub> in atmosphere (LC<sub>25</sub>, LC<sub>40</sub> and LC<sub>50</sub>) on the fecundity of *C. maculatus* when the two sexes were exposed for 3 days, revealed that the LC<sub>25</sub> did not highly affect fecundity. At LC<sub>40</sub> and LC<sub>50</sub> levels, the number of eggs laid per female was greatly reduced. Data showed that the sublethal concentrations of CO<sub>2</sub> in the atmosphere did not inhibit egg laying. These results are in agreement with those of Press and Flaherty (1973) who found a large reduction in the fecundity of *Ephestia Kuhnella* (Zeller) *Ephestia cautella* (Walker) and *Plodia interpunctella* (Hubner) when exposed to 96% CO<sub>2</sub> in atmosphere for 2 h per day.

### Effect on egg hatchability

The hatchability of eggs laid by the progeny of all treatments was generally less than that of the untreated females (Fig. 2). The percentages of hatchability were 78.5 %, 71.1 % and 59.3 % for LC<sub>25</sub>, LC<sub>40</sub> and LC<sub>50</sub>, respectively, compared with 80.6 % for the untreated control. Hatchability was much more affected in treatment with LC<sub>50</sub> compared with other treatments.

### Effect on longevity

At the LC<sub>50</sub> level, the adults lived 3 days shorter than the untreated adults (Fig. 3). However, decreasing the concentration to LC<sub>25</sub> had resulted in one day shortening in adult's life span. In general, the longevity of females at all treatments was slightly shorter than males. Storey (1975) on his studies on the developmental period and longevity of *C. maculatus*, *Sitophilus oryzae* (L.) and *Sitophilus granarius* (L.) came to the same conclusion.

### Effect on reproductive capacity

As shown in Fig.4, the reproductive potential of *C. maculatus* was considerably reduced by increasing the sublethal concentration from LC<sub>25</sub> to LC<sub>50</sub>.



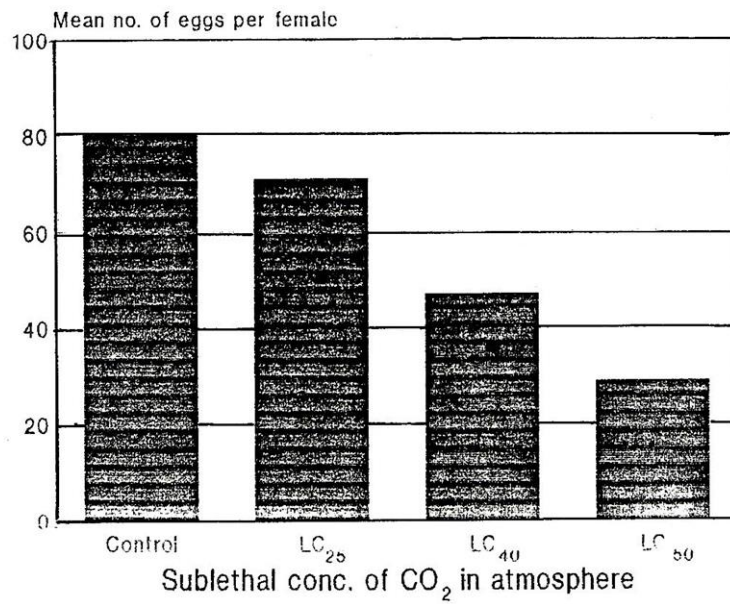


Fig. 1. Effect of sublethal concentrations of CO<sub>2</sub> in atmosphere on the fecundity of the survivor females of *C. maculatus*.

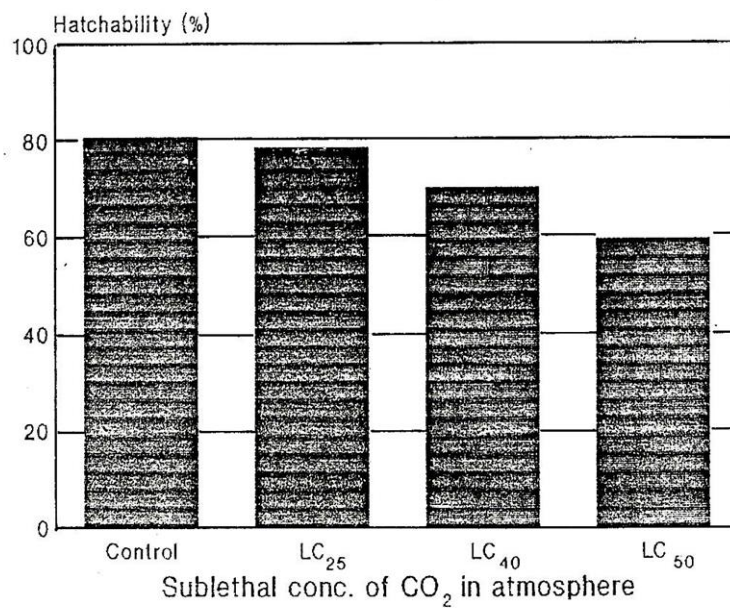


Fig. 2. Effect of sublethal concentrations of CO<sub>2</sub> in atmosphere on the hatchability (%) of the survivor females of *C. maculatus*.

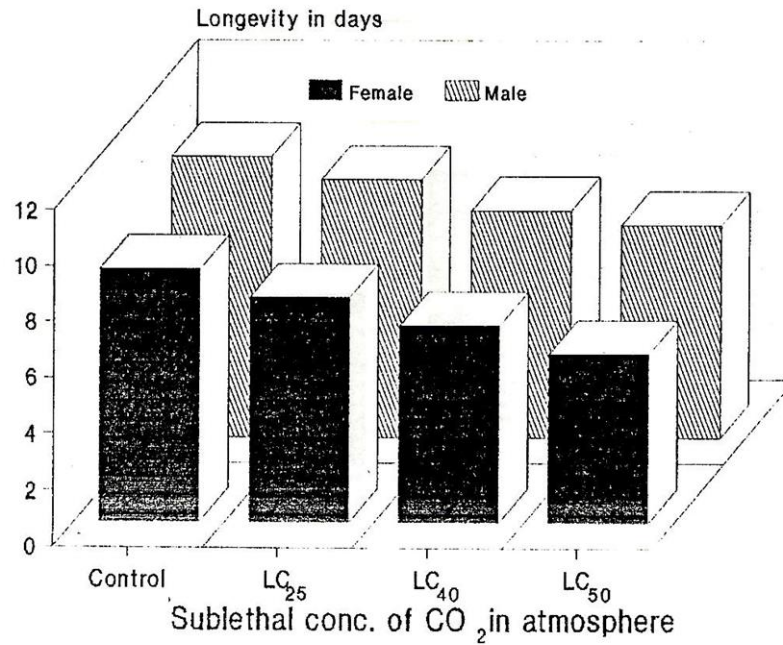


Fig. 3. Effect of sublethal concentrations of CO<sub>2</sub> in atmosphere on the longevity of the survivor females and males of *C. maculatus*.

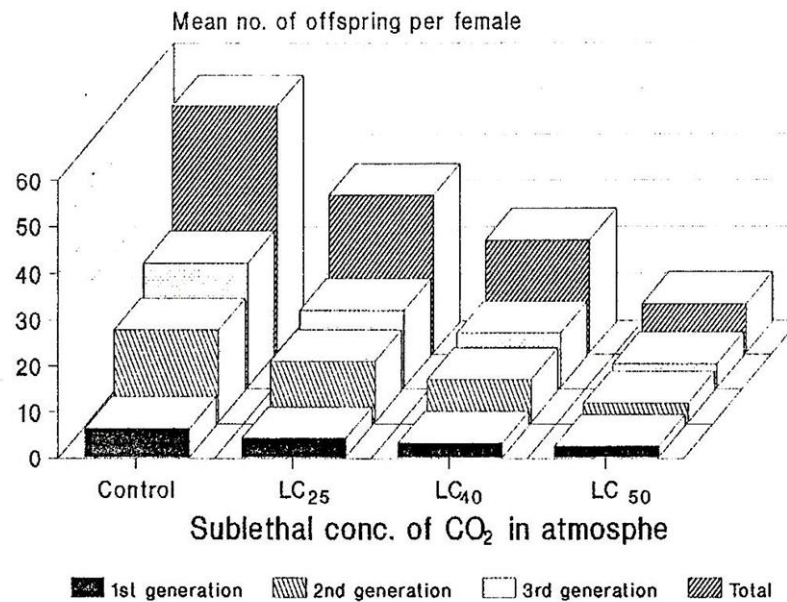


Fig. 4. Effect of sublethal concentrations of CO<sub>2</sub> in atmosphere on the mean no. of offspring of *C. maculatus* emerged after three generations per female.

The mean number of offspring produced per female was 63.8, 45.4 and 38.5% of the number produced by untreated female after three generations under LC<sub>25</sub>, LC<sub>40</sub> and LC<sub>50</sub>, respectively. This significant reduction in the reproductive potential agree with the findings of Spratt (1979a) on *Sitophilus zeamais*.

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## تأثير التركيزات تحت المميته لغازك أ في مخلوط الهواء على السلوك البيولوجي لحشرة خنفساء اللوبيا

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أجريت دراسات على التركيزات تحت المميته لغازك أ في مخلوط الهواء ( LC50, LC25, LC40 ) وتأثيرها على السلوك البيولوجي لحشرة خنفساء اللوبيا. وقد اتضح أنه عند كل التركيزات تحت المميته للغاز تقصر فترة الحياه لإناث الحشرات الكامله عنها فى حالة الذكور. وأنه بزيادة التركيزات التحت مميته للغاز تقصر فترة الحياه للحشرات الكامله. وكذلك بزيادة تركيزات الغاز التحت مميته فإن عدد البيض الموضوع بالنسبه للحشرة الأنثى ينخفض بدرجة كبيره ولكن هذه التركيزات لا تمنع وضع البيض.

وقد ظهر واضحاً أن زيادة التركيز التحت مميت للغاز فى الهواء إلى LC50 قد قلل بدرجة ملحوظه من خصوبة الحشرات الكامله (بقياس متوسط عدد الخلفه الناتجه من حشره أنثى واحدة).

هذه الدراسات قد أجريت على خلفه ناتجه من بيض سبق تعريضه للتركيزات التحت مميته المذكوره سابقاً لمدة تعريض قدرها ٣ أيام.