

## Effects of plant oils on oviposition preference and larval survivorship of *Callosobruchus chinensis* (Coleoptera: Bruchidae) on azuki bean

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### Abstract

The efficacy of four kinds of oils, neem, castor, sunflower and sesame, was evaluated for the control of infestation of stored azuki bean with *Callosobruchus chinensis*. Neem and sesame oil-treated beans were less preferred and oviposition of adults on these beans decreased. After three days of release, 100% adults were found dead on neem oil-treated beans and very few eggs were laid. Effects on the survival rate of immature beetles and percentage of seed weight loss were also recorded. Neem and sesame oils completely inhibited adult emergence and appeared to be most promising as a seed protectant against *C. chinensis*. The oil-specific activity and delay in larval development in oil-treated beans suggest that the protective properties involved chemical rather than physical factors.

**Key words:** *Callosobruchus chinensis*, plant oils, Bruchidae, storage pest control, *Vigna angularis*

### INTRODUCTION

Azuki bean, *Vigna angularis*, which is an important edible bean in Japan is prone to infestation with various insect pests. Among these, the azuki bean weevil, *Callosobruchus chinensis*, is the major pest in storage as well as in the field (Shinoda and Yoshida, 1989). Gujar and Yadav (1978) recorded 55-60% weight loss of stored Azuki beans and 46-66% loss in protein content by infestation with the beetles. Pesticides are mainly used for successful storage, although these chemicals are hazardous to human health and the environment. Thus, the practice of mixing them with grain is discouraged.

Use of various kinds of plant materials and oils for the control of storage pests is being implemented in many parts of the world. Mixing of different kinds of oils and plant materials (leaf, bark, seed, etc.) was traditionally practiced especially in the southern part of Asia and in Africa. A large number of studies have been carried out on the effectiveness of different kinds of vegetable oils against *Callosobruchus* spp. It was reported that some oils inhibited multiplication of the beetle populations (Sangappa, 1977; Ali et al., 1983; Ahmed et al.,

1993). However, there is little information about the critical life stage when the beetle population experiences a decrease, and also the mechanisms of the decrease are unclear (Pacheco et al., 1995). In particular, no studies have been carried out on the oviposition preference of adult beetles among beans treated with different kinds of oils.

As a first step to analyze the mechanism of the oil effect on the protection against *C. chinensis*, in this paper, the variation of oviposition preference of adults and mortality of immature forms among beans that were treated with four different plant oils were investigated.

### MATERIALS AND METHODS

**Rearing of test insects.** The insect pests, *C. chinensis* were collected from cultivated Azuki beans in Miki-cho, Kagawa, Japan and maintained in the laboratory on Azuki beans at 25°C, under a 70±5% R.H. and 14L : 10D regime. Azuki beans grown and harvested in Hokkaido, Japan were used for the rearing. To avoid preinfestation, the beans were kept in the oven at 40°C overnight, then placed in a large glass jar covered with a netted cloth lid for one day to let them acquire normal seed moisture without further infestation.

**Oils.** In this experiment four kinds of oils, neem, *Azadirachta indica*, castor, *Ricinus communis*, sunflower, *Helianthus annas* and sesame, *Sesamum indicum* were used. The oils were all crude and not refined.

**Effect on preference.** For each treatment 50 fresh beans were weighed and placed in a petri dish 5.5 cm in diameter. Oil (v/w) was added at 0.5% to each petri dish, which was then shaken to obtain an even distribution of oils on the surface of the beans. Four petri dishes, each containing beans treated with different kinds of oils and a control (a petri dish with 50 untreated beans) were kept open in a large tray 35 cm in diameter in which the five petri dishes were arranged in a circular manner. Then in the center of the tray 20 pairs of 1-day old beetles were carefully introduced, and the large tray was covered with a lid. Five replications were made. The beetles were allowed to lay eggs freely for three days, and then the numbers of adults and deposited eggs in each petri dish were counted.

**Effect on survivorship.** Emergence of adult progeny was recorded daily during a period of 90 days. The percentage of weight loss of the beans was also recorded after 90 days of adult release. All the experiments were conducted in a chamber controlled under the following conditions: 25°C, 70±5% R.H. and 14L : 10D regime.

## RESULTS AND DISCUSSION

### Effect on preference

The mean number of adults (dead plus alive) in each petri dish ranged from 2.0 to 12.6 (Table 1). The lowest number of adults was detected in the neem oil-treated petri dish where 100% of the insects were found dead. The highest number of adults was found in the control. In the control, 100% of the insects were found alive whereas 66, 34 and 10% of the adults were found alive in castor, sunflower and sesame oils, respectively (Table 1). These results are in agreement with those reported by Dohary et al. (1988) whereby a high dose (1 ml/100 g) of soybean, neem and sesame oils caused 100% adult mortality of *C. chinensis* and *C. maculatus* after one day of exposure.

The mean numbers of eggs laid were significantly different between the treatments (Ta-

Table 1. Number of adult beetles (% alive) and deposited eggs (mean±S.E.) in each petri dish containing oil-treated beans.<sup>a</sup> Control beans were not treated with oils.

Treatment	No. of adults (% alive)	No. of eggs <sup>b</sup>
Control	12.6±0.9 a (100.0)	403.2±7.1 a
Castor	6.4±0.6 b (65.8)	181.8±36.6 b
Sunflower	7.0±1.3 b (33.9)	47.2±15.2 b
Sesame	5.6±0.7 bc (9.5)	17.8±7.8 bc
Neem	2.0±0.5 c (0.0)	2.4±0.5 c

<sup>a</sup> Values followed by the same letters in the same column did not differ significantly by Tukey's test ( $p < 0.05$ ).

<sup>b</sup> Number of eggs ( $Y$ ) was transformed to  $\log(Y+1)$  and subjected to the test.

ble 1). The lowest number of eggs (2.4) was laid on neem oil-treated beans, followed by sesame (17.8), sunflower (47.2) and castor (181.8) oil-treated ones. The highest number of eggs was found in the control (403.2), which was significantly different from the others. These results, together with similar findings reported by Khaire et al. (1992), indicate that the oviposition is inhibited when stored beans are treated with some kinds of oils. Furthermore, our results suggest that not only the reluctance of adults to visit and oviposit on some oil-treated beans, but also the higher adult mortality on those beans contributed to the lower oviposition rate. Based on these results we propose three mechanisms for the prevention of adult oviposition on oil-treated beans. 1. Some kinds of volatile substances in the oils prevent adult access to the oil-treated beans; 2. Some kinds of toxic components in oils kill adults on the

Table 2. Survival rate of *C. chinensis* from egg to adult in oil-treated azuki beans and percentage of weight loss of the beans after 90 days<sup>a</sup>

Treatment	%Survival	%Weight loss
Control	85.4±2.4 a	62.0±0.9 a
Castor	9.7±4.0 b	8.2±2.2 b
Sunflower	0.4±0.4 c	4.5±0.3 b
Sesame	0.0±0.0 c	4.5±0.3 b
Neem	0.0±0.0 c	4.0±0.7 b

<sup>a</sup> Values are mean±S.E. Data were transformed to arcsine and subjected to Tukey's test. Means followed by the same letters in the same column did not differ significantly at  $p < 0.05$ .

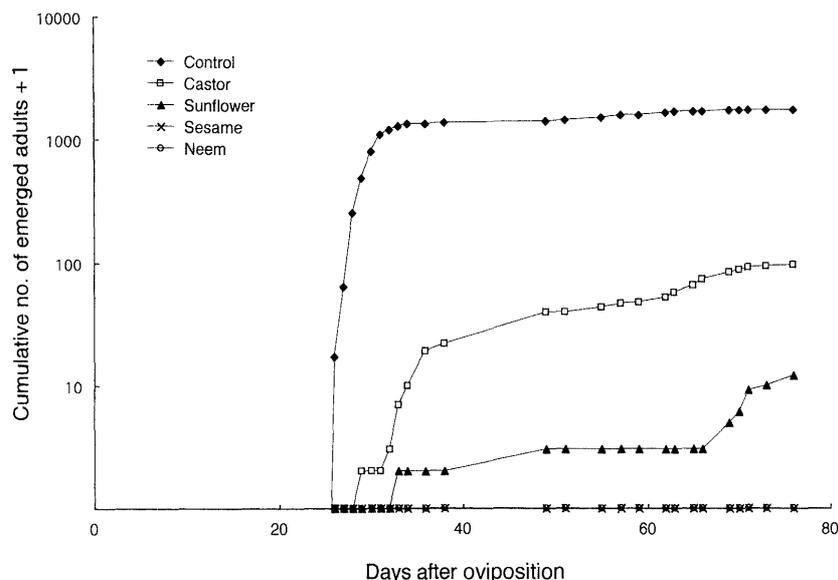


Fig. 1. Cumulative number of adult beetles that emerged from oil-treated and untreated beans.

beans; and/or 3. Mechanical or chemical properties of oils inhibit the oviposition behaviour of adults.

#### Effect on survivorship

Percentage of adult emergence ranged from 0 to 85.4 (Table 2). The highest percentage of adult emergence occurred in the control, in contrast with the other treatments. Neem and sesame oils completely inhibited adult emergence while sunflower and castor oils also prevented normal survivorship. In the control, the adults started to emerge after 26 days of oviposition and the rate of adult emergence was high from 27 to 35 days after oviposition (Fig. 1), whereas in castor oil-treated beans the adults started to emerge after 32 days of oviposition and the rate of emergence was high from 34 to 38 days after oviposition. In the case of sunflower oil-treated beans, the rate of adult emergence was high after 70 days of oviposition. These results suggest that oils not only decrease the survival rate but also prolong the developmental period.

The mean percentage of bean weight loss 90 days after the beetle release ranged from 4.0 to 62.0 in oil-treated and untreated beans (Table 2). Compared with the control, all the treatments showed a lower seed weight loss because of the lower insect infestation.

Overall, such low performances of immature beetles in oil-treated beans suggest two possi-

bilities: 1. The eggs on the seed coat are killed due to the interference of oils with respiration (physical protection); and/or 2. Toxic substances in oils kill the immature beetles inside the beans (chemical protection). The oil-specific activity and the delay in larval development suggest that the second hypothesis is more plausible.

Considering the efficacy of different kinds of oils in terms of adult mortality, oviposition rate, developmental mortality and seed weight loss, neem and sesame oils appeared to be most promising as seed protectants. To elucidate the mechanism(s) responsible for such lower preference and performance of bruchids on oil-treated beans, further experiments should be carried out.

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