



OVIPOSITION PREFERENCE OF *CALLOSBRUCHUS MACULATUS* (F.) TO COMMON PULSES AND POTENTIALITY OF TRIFLUMURON AS THEIR PROTECTANT

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Abstract

Seeds of black grams, lentils, Bengal grams and green peas were soaked separately in aqueous solutions of Triflumuron at doses of 0.0 (control), 0.5, 1.0, 1.5 and 2.0 ppm. Three day-old adults of *Callosobruchus maculatus* (F.) (Coleoptera: Bruchidae) were allowed to oviposit on air-dried, treated or untreated pulses of each type and dose in 'no-choice' and 'free-choice' tests. Maximum oviposition occurred in Bengal grams (61.00±0.25) and green peas (33.67±0.54) in 'no-choice' and 'free-choice' tests, respectively. The beetles avoided egg laying on lentils. Maximum egg-hatching occurred on black grams (>90%) in both tests. Minimum developmental time was required in the Bengal grams (30±0.5 days in 'no-choice' test and 31.33±1.23 days in 'free-choice' test). No adults emerged from green peas, while 54.79% and 86.15% emergence were recorded from untreated black grams in 'no-choice' and 'free-choice' tests, respectively. Triflumuron reduced egg-laying significantly in green peas, where hatchability also reduced significantly to 35.99% at 2 ppm in 'no-choice' test. Percentage of hatching decreased in all pulses with the increasing doses of triflumuron. No adults emerged from the treated green peas in any test, and at 2 ppm the adult emergence declined to <50% in all pulses. Implications of these results are further discussed.

Key words: *Callosobruchus maculatus*, Triflumuron, seed protectant, fecundity, hatchability, developmental period, adult emergence

Introduction

The pulse beetles *Callosobruchus* (Coleoptera: Bruchidae) are the major pests of pulses in Bangladesh (Begum *et al.* 1993). A single beetle could cause 3.5% weight loss to cowpea seeds (Booker 1967). Gujar and Yadav (1978) recorded 55 to 60% loss in seed weight and 45.50 to 66.30% loss in protein content of pulses due to infestation caused by these beetles. Infestation of *Callosobruchus* mainly begins in the field, where the beetles lay eggs on the mature pods of the pulses. The immature stages are internal feeder, causing a total damage to the pulse seeds.

Because of the increasing threats of the conventional insecticides to environment and human beings, alternative pest control measures are being searched throughout the globe. These include, among others, the use of such insect growth regulators (IGRs) as diflubenzuron and triflumuron (Haynes and Smith 1989, Parween 1996a), vegetable oils (Ali *et al.* 1983, Bhaduri *et al.* 1990), natural pesticides from neem tree (Schmutterer 1990) and repellents like pulse flour (Fields *et al.* 2001, Kumar *et al.* 2004). Previous studies showed that triflumuron not only retards larval development (Mian and Mulla 1982, Eisa *et al.* 1984, Elek 1994), but it also reduces egg-laying and fertility of the adults in a wide range of stored products insects (Mian and Mulla 1982, Eisa *et al.* 1984, Elek and Longstaff 1994, Parween 1996b). Kumar *et al.* (2004)

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evaluated the repellent effects of the whole-pea flour against the stored grain insects like the red flour beetle, the rice weevil and the lesser grain moth. In the present study triflumuron was tested for its efficacy as a protectant against the pulse beetle reared on four common pulse seeds.

Materials and Methods

Insects

Newly emerged adults of *C. maculatus* were used for evaluating their oviposition preference on untreated and triflumuron-treated seeds in two sets of experiments detailed below.

Treatments

Required quantities of Triflumuron (common name is Baycidal which is a product of Bayer AG, and contains 25% wettable powder of triflumuron), were dissolved in distilled water and shaken well to obtain doses of 0.0 (control), 0.5, 1.0, 1.5 and 2.0 ppm. Good quality pulses were used after sterilizing at 60°C in an oven. Fifty seeds of each pulse species were soaked separately in each dose for 30 minutes. The seeds were then removed from the solution and dried in air at room temperatures ranging from 25 to 30°C.

Experiments

Two experiments were set up on the basis of 'no-choice' and 'free-choice' of the beetles on four different pulse species viz, black grams (*Vigna mungo*), lentils (*Lens esculentus*), Bengal grams (*Cicer arietinum*) and green peas (*Pisum sativum*) in all five doses. The experiments were conducted at room temperatures (25-30°C).

No-choice' tests

Fifty seeds of each of the four pulse species were placed separately in 9-cm diameter Petri dishes. The seeds were arranged in a single layer without any overlapping. Single pairs of 3 day-old adult *C. maculatus* males and females were released in each Petri dish for egg laying. The beetles were removed after 48 hours. The number of eggs laid, egg-hatchability (%), egg-to-adult developmental period (days) and adult emergence (%) from each pulse seeds were recorded.

Free-choice' tests

Fifty seeds of each of the pulse species were placed in a Petri dish (15-cm diameter). Single pairs of 3 day-old adult males and females of *C. maculatus* were released in the Petri dish and removed after 48 hours. The beetles had unrestricted access to all pulses for egg-laying. Data were recorded as in 'no-choice' tests.

Statistical analysis

The data were subjected to two-way ANOVA to evaluate three possible effects: between pulses, between triflumuron doses, and interactions between pulses and doses. The mean values of the experiments were separated using Duncan's Multiple Range Test (DMRT) following procedures of Gomez and Gomez (1984).

Results and Discussion

Preference of *C. maculatus* to untreated pulse seeds

Given restricted access to only one type of pulse seeds ('no-choice' tests), *C. maculatus* preferred Bengal grams (61.00±0.25) to black grams (33.67±0.70), green peas (30.67±0.67) and lentils (21.33±0.12) for oviposition (Table 1). Under unrestricted access to pulses ('free-choice' tests), the beetles preferred egg-laying on green peas (33.67±0.54) > black grams (25.67±0.25) > Bengal grams (10.67±0.49) > lentils (2.00±0.01). The maximum hatching of eggs occurred in black grams (92.18%) followed by Bengal grams (75.56%), green peas (74.24%) and lentils (54.59%) in restricted access to pulses. A similar trend was also observed for the unrestricted option (Table 1). The egg-to-adult development took 30.00±0.50 days in

Bengal grams, 35.09±0.17 days in black grams and 37.44±0.11 days in lentils, whereas green peas did not support immature development of the beetles in both choice conditions (Figs.1a and 1b). The maximum adult emergence in no-choice tests took place in black grams (54.79%), as compared with Bengal grams (29.37%), lentils (17.59%) and green peas (0.00%) while under 'free-choice options, the adult emergence enhanced to 86.15%, 33.33% and 27.87% in black grams, Bengal grams and lentils, respectively, but no adults emerged from green peas (Figs. 2a and 2b).

Table 1. Triflumuron-induced changes in fecundity and egg-hatch on different pulse seeds in the 'no-choice' and 'free-choice' tests in *C. maculatus*

Pulses/Doses (ppm)	'No-choice' test		'Free-choice' test	
	Fecundity ¹ (Mean±SD)	Egg-hatch ² (%)	Fecundity ³ (Mean±SD)	Egg-hatch ⁴ (%)
Black grams				
0.0 (control)	33.67±0.70a	92.18a	25.67±0.25a	95.45a
0.5	31.67±0.25a	67.97b	17.67±.70b	59.52b
1.0	31.67±0.41a	64.54b	17.33±0.25b	56.06b
1.5	38.33±0.41a	59.58c	18.67±0.22b	40.35c
2.0	35.00±0.50a	59.55c	14.33±0.05b	27.15d
Lentils				
0.0 (control)	21.33±0.12a	54.49a	2.00±0.01	100.00
0.5	26.00±0.20a	57.35a	0.00	0.00
1.0	20.33±0.14a	57.96a	0.00	0.00
1.5	25.67±0.70a	53.09a	0.00	0.00
2.0	26.67±0.71a	32.22a	0.00	0.00
Bengal grams				
0.0 (control)	61.00±0.25a	75.56a	10.67±0.49a	33.33a
0.5	43.01±0.21b	85.02b	11.67±0.90a	30.56a
1.0	37.33±0.25b	81.81b	7.68±0.80b	28.57a
1.5	26.34±0.70c	58.42c	2.35±0.20c	25.00b
2.0	21.05±0.84c	47.44d	3.00±0.50c	24.37b
Green peas				
0.0 (control)	30.67±0.67a	74.24a	33.67±0.54a	93.03a
0.5	31.67±0.55a	63.03ab	39.64±0.23b	87.28a
1.0	25.33±0.74b	62.79ab	37.67±0.22b	76.88b
1.5	23.67±0.44b	56.02b	29.33±0.14a	54.76c
2.0	23.00±0.50b	35.99c	19.01±0.74c	38.01d

¹F_{pulses}= 6.55 P<0.01; F_{doses}= 3.36, P<0.05; F_{pulses×doses}= 1.90ns; ²F_{pulses}= 1.25ns; F_{doses}= 1.70ns; F_{pulses×doses}= 1.09ns; ³F_{pulses}= 53.69 P<0.001; F_{doses}= 1.03ns; F_{pulses×doses}= 1.54ns; ⁴F_{pulses}= 5.43 P<0.01; F_{doses}= 3.53, P<0.05; F_{pulses×doses}= 1.70ns. Means followed by the same letters in a column in each pulse and test are not significantly different by DMRT at P<0.05; ns = not significant.

Fig. 1a. Effect of triflumuron on developmental period on different pulse seeds in 'no-choice' test in *C. maculatus*

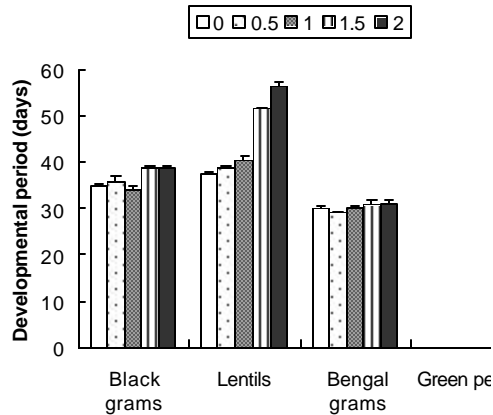


Fig. 1b. Effect of triflumuron on developmental period on different pulse seeds in 'free-choice' test in *C. maculatus*

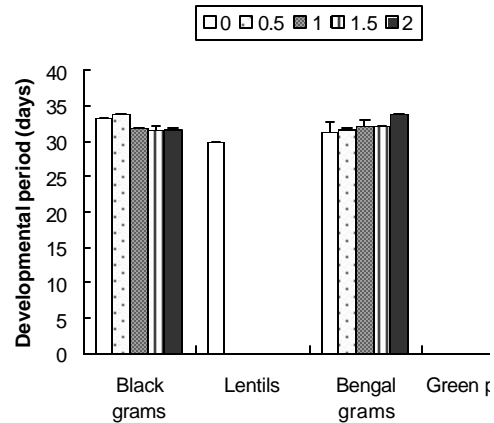


Fig. 2a. Effect of triflumuron on adult emergence from different pulse seeds in the no-choice test in *C. maculatus*

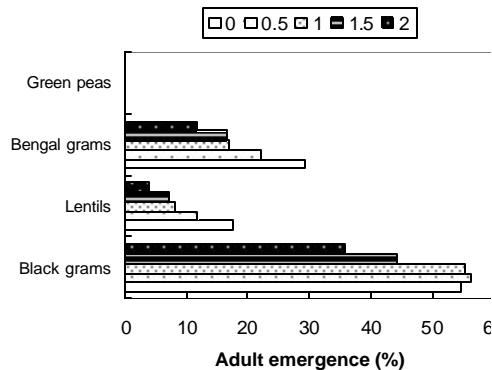
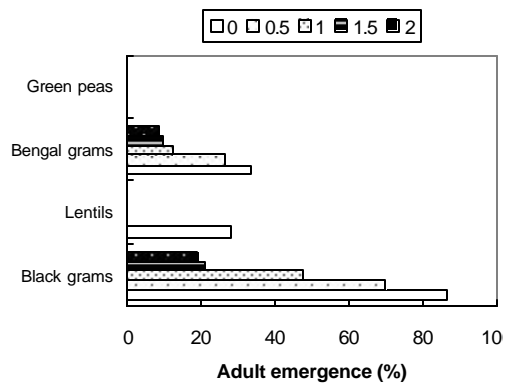


Fig. 2b. Effect of triflumuron on adult emergence from different pulse seeds in the 'free-choice' test in *C. maculatus*



Potentiality of triflumuron against *C. maculatus* infestation

Triflumuron-induced changes in the reproductive parameters of *C. maculatus* are shown in Table 1 and Figs. 1-2. Compared to controls, significant reductions in egg-laying occurred in Bengal grams (21.05±0.84) and green peas (23.00±0.50) under no-choice situations. But interestingly, oviposition in 'free-choice' options reduced significantly in green peas (19.01±0.74), black grams (14.33±0.50) and Bengal grams (3.00±0.5), while the beetles completely avoided egg-laying on lentils. Egg-hatching was significantly reduced in black grams (59.55%) followed by Bengal grams (47.44%), green peas (35.99%) and lentils (32.22%) in restricted access. Reductions in hatchability also took place in unrestricted access to pulses: green peas (38.01%) > black grams (27.15%) > Bengal grams (24.37%) > lentils (0.00%). In 'no-choice' tests triflumuron treatments did not affect immature developmental period in black grams (38.78±0.5 days) and Bengal grams (31.11±0.8

days), but they significantly lengthened it in lentils (56.22±0.9 days) and immatures failed to develop in green peas. Similar results were observed in 'free-choice' options except that development did not proceed in lentils and green peas. Significantly reduced adult emergence in black grams (36.02%), Bengal grams (11.61%) and lentils (3.7%) resulted following triflumuron treatments in 'no-choice' tests. Further reduction in adult emergence occurred in black grams (19.05%) and Bengal grams (8.33%) in 'free-choice' situations where no adults emerged from lentils and green peas.

It is obvious from the present results that *C. maculatus* has preference for pulse seeds. It preferred Bengal grams and green peas to black grams and lentils under compulsive (*i.e.* 'no-choice') and unrestrictive (*i.e.* 'free-choice') situations for egg-laying. Chemical cues and/or textures of the seed coat might be the reason for such differential choices. Green peas did not support immature development, and lentils had been the least preferred pulse for oviposition in the experimental *C. maculatus*. This lends support from previous work by Teotia and Singh (1966), Shazali (1989) and Begum *et al.* (1993). In addition, an earlier report that pea-flour contains repellent agents and growth deterrent for a number of stored product insects (Kumar *et al.* 2004), is also supportive to the present results. Black gram not only allowed maximum egg-hatch but it also supported maximum number of the beetles to complete their metamorphosis which is similar to findings by Mannan and Bhuiyah (1996) where hatchability of *C. maculatus* in cowpeas was over 85%. Compared to black grams and lentils, however, the egg-to-adult developmental period in Bengal grams was shorter, the reason of which is not clear yet.

Triflumuron effectively reduces reproductive potential in a number of stored product pest insects (Mondal and Parween 2000). Although triflumuron is mainly a stomach poison, it also has some contact action against the eggs (Fox 1990). This caused reduced hatching of the eggs in *C. maculatus*, though egg-laying by the females remained unaffected on some of the treated pulses. Higher doses and longer exposure times of triflumuron might be required to further reduce fecundity of *C. maculatus* on treated pulses. Unlike Bengal grams and green peas where higher doses of triflumuron resulted in reduced egg-laying, oviposition was not deterred on black grams and lentils by triflumuron treatments. This perhaps reflects the fact that IGRs might act as oviposition inducer for certain stored product insects (Mian and Mulla 1982). Akhter *et al.* (2003) reported that though triflumuron did not affect parental fecundity, it significantly reduced fecundity in the F₁ progenies when treated beetles were crossed with untreated counterparts. But it should be kept in mind that triflumuron inhibits chitin synthesis, it results in the cessation of feeding (Parween 1996a) and disruption of the mid-gut structures, thus causing interference with digestion and absorption (Parween 1997). It could also be capable of inducing partial or complete sterility in the experimental beetles as were the cases with *Tribolium castaneum* (Parween 2003, 2004) and *Alphitobius diaperinus* (Begum *et al.* 2003). The present results therefore are suggestive of using carefully estimated doses of triflumuron as a surface protectant for pulse seeds against *Callosobruchus* infestation.

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