Effect of Triflumuron on the development of the red flour beetle, *Tribolium castaneum* Herbst (Coleoptera: Tenebrionidae)

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Abstract

The effects of triflumuron on the development of *Tribolium castaneum* Herbst were studied Larval treament of 2.5×10^{-7}, 2.5×10^{-8} and 2.5×10^{-9} ppm of the compound for 16 days significantly ($P<0.01$) reduced the weight of larvae, pupae and adults, and prolonged the developmental periods. At a dose of 2.5×10^{-9} ppm normal pupal formation and adult emergences were reduced significantly ($P<0.01$) due to deformities, while at 2.5×10^{-7} and 2.5×10^{-8} ppm it was due to higher larval mortality. Sex identification was not possible in the pupae emerged from the triflumuron treated medium at 2.5×10^{-7} ppm, and in other two doses it was possible and the number of males was 1.5 and 2 times higher than the number of females, respectively. In treated media egg mortality was decreased with the age of eggs, and was increased with the exposure time. There was no relationship between the percentage of egg mortality and the doses, and nearly 50% mortality occurred at each experiment. Triflumuron was also found to reduce adult population of *T. castaneum* in three months exposure time.

Introduction

Insect growth regulators are (IGRs) gaining importance over the conventional neurotoxic chemical pesticides used in food stores against insect pests, mainly because they have low mammalian toxicity and degrade rapidly in the environment (Antognini, 1972; Eisler, 1992). The benzoylphenyl urea (BPU) compounds, a group of IGRs, were found to inhibit chitin synthesis in insects (Post and Vincent, 1973; Post et al., 1974; Deul et al., 1978; Gijswijt et al., 1979) and effective larvicides (Mulder and Gijswijt, 1973; Ishaaya and Casida, 1974; Grosscurt, 1978; Hammock and Quistad, 1981) at low doses (Loschiavo, 1976; Mian and Mulla, 1982a, b; Eisa et al., 1984; Elek, 1994). Generally, BPUs do not kill adult insects but prevent juvenile stages from completing their development (Dales et al., 1994).

Triflumuron (code name: Bay SIR 8514, commercial name Baycidal), a product of Bayer, is a promising larvicide effective at very low dose, against the insect pests both in the field and stores. The compound inhibits chitin synthesis and controls growth and development of various insect species of stored commodities (Kramer and McGregor, 1979; Ishaaya et al., 1981, 1986; Mian and Mulla 1982a, b; Eisa et al., 1984, 1986; Faragalla et al., 1985; Elek and Longstaff, 1994).

Tribolium infestation is a worldwide major problem in stored products and food factories, resulting in both contamination and substantial economic damage (Mondal, 1994; Mondal and Port, 1994). The present work aims to determine the effectiveness of triflumuron on the development of the red flour beetle, *Tribolium castaneum* Herbst.

Materials and Methods

Triflumuron

The compound Baycidal, containing 25% of triflumuron, a disubstituted benzoylphenyl urea compound, was collected from Bayer AG, Germany. The compound, a wettable powder, was added in appropriate quantities to the flour medium to obtain doses of 2.5×10^{-7}, 2.5×10^{-8} and 2.5×10^{-9} ppm, which were used in the presented experiments

Insect

T. castaneum were obtained from the laboratory culture of the Institute of Biological Sciences, Rajshahi University, Bangladesh. The beetles were maintained on wheat flour at 30℃ without controlling light and humidity.

Larval growth

Newly hatched larvae were kept in glass vials (50×25cm)

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containing approximately 0.5g of food either treated or untreated. The tops of the vials were secured by cotton wool and kept in an incubator at 30℃. The food medium was replaced every fourth day to avoid conditioning by the larvae themelves (Mondal, 1983) Larave were regularly observed until pupation and the larval period was noted. Larval weight was registered on the 10th and 16th day from hatching, though larval instars in the treated medium was not known, but they corresponded to the 3rd and 6th instars of the control larave, respectively (Mondal, 1984). Weight of the emerged pupae was taken on the 3rd day after pupation

Adult emergence and sex-ratio

Three hundred newly hatched larvae were introduced in each treatment in 1 1b Kilner jars The larvae were checked regularly for pupation. The pupae were separated from the jar and the normal pupae were sexed by observing the exogenital process of females(Halstead, 1963). The sexed pupae were kept separately in petri dishes for adult emergence, and the pupal period was noted. Each experiment was replicated thrice the control was maintained in the same way with the untreated medium.

Abnormalities

Abnormal larvae, pupae and adults were separated from treated media. Abnormalities were recorded and the percentage of deformities was determined.

Egg mortality

Batches of 300 eggs, 0 − 24, 24 − 48, 48 − 72 and 72 − 96 hrs old respectively, were kept in food medium treated with aforementioned doses of triflumuron, for 24, 48 and 144 hrs Eggs exposed for 24 and 48 hrs were separated from the treated medium and kept on an untreated filter paper. The same number of eggs was kept in untreated medium as control. All the treatments were replicated thrice.

Adult population

Thirty unmatd eight days old pairs of adults were kept in a 1 1b glass jar containing 100g of food medium either treated or untreated with triflumuron of the mentioned doses. After 30 days each, and additional amount of 50g of treated or untreated food medium was added to each jar to reduce both conditioning and overcrowding in the food (Mondal and Port, 1995). The total number of adults was assessed after 90 days. The percent reduction in adult population was calculated using the formula:

Percent reduction in adult progeny $= (1 - t/c)100$

Where t = number of adults in the treated food,

c = number of adults in the untreated food

Results

Table 1 shows that triflumuron at doses of 2.5×10^{-7}, 2.5×10^{-8} and 2.5×10^{-9} ppm reduced the larval and pupal weight ($P<0.01$) and lengthened both larval and pupal developmental periods($P<0.01$) of *T. castaneum*.

The sex-ratios of the beetles at the doses 2.5×10^{-8} and 2.5×10^{-9} ppm were deviated from 1 1 with 1.5 and 2 times higher number of males than the females, respectively (Table 2). At the highest dose (2.5×10^{-7} ppm) most of the pupae were deformed remaining within the larval exuvae and hence sex identification was not possible, but the sex ratio did not deviate in normal pupae. Percentages of pupal and adult emergence were significantly reduced ($P<0.01$) in the treated larvae (Table 2).

Larval mortality was higher at the doses of 2.5×10^{-7} and 2.5×10^{-8} ppm, but at the lowest dose (2.5×10^{-9} ppm) abnormalities were comparatively higher than at the other two doses, for all the stages (Table 3) Larval-pupal and pupal-adult intermediate forms were common at each dose. The larval treatment produced higher number of adult deformities (Talbe 3) showing various morphological deformed characters (Table 4). The intermediate forms survived for a short period

Triflumuron caused about 50% mortality of the *T castaneum* eggs (Table 5). Both the age of eggs or the exposure period did not have any effect on the egg mortality.

A three months exposure of adult beetles to triflumuron reduced the adult population (Table 2). The percent reduction of adult population were 59.71, 31.19 and 16.87 at doses of 2.5×10^{-7}, 2.5×10^{-8} and 2.5×10^{-9} ppm, respectively.

Discussion

Triflumuron and related compounds need longer exposure of larvae to show lethal effects (Ishaaya et al., 1986; Ishaaya and Yablonskii, 1987; Parween, 1996). Prolonged larval exposure to media treated with low dose of BPUs, affects development due to the imparied cuticle and failure to ecdyse. In the present study the survivability of *T. castaneum* larvae was severely affected by triflumuron at dose of 2.5×10^{-7} and 2.5×10^{-8} ppm. At the exposure of 20 days only 60% pupal eclosion was observed at a dose of 2.5×10^{-9} ppm, but about 96% of the larvae pupated in control (Table 2). Weights of the treated larvae and pupae were reduced by the treatment. BPU compounds have been reported to inhibit growth in various insects by reducing larval and pupal weight Reduced larval weight was reported in diflubenzuron and triflumuron treated *T. confusum* (Ishaaya et al., 1984), diflubenzuron, CME-134, XRD-473 and IKI-7899 treated susceptible and multi-resistant strains of *T. castaneum* (Ishaaya et al., 1986; Ishaaya and Yablonskii, 1987) and triflumuron treated suceptible and multi-resistant strains of *T. castaneum* (Parween, 1996).

Due to the interference with cuticle formation the developmental period of the treated larvae and pupae were prolonged at all doses of triflumuron. Similarly, triflumuron (0.05 ppm) increased the larval period in *T. confusum* (El-Sayed et al , 1984 – 1985) and diflubenzuron (10 ppm) in *Trogoderma granarium* (Saxena and Kumar, 1991). Triflumuron at 1×10^{-7} ppm prolonged the larval period of both susceptible and resistant strains of *T castaneum* (Parween, 1996). Elek et al. (1990) and Estal (1994) reported slightly longer developmental periods in *T. castaneum* and *Rhyzopertha dominica* when the larvae were treated with low doses of BPUs including triflumuron. Longer pupal period was also reported in diflubenzuron treated *T. granarium* by Soltani et al. (1984). Triflumuron in the food media acts as an antifeedant (Parween, 1997), which may be the cause of retard growth of *T castaneum* larvae, as found in the present study.

Table 1. The mean larval weight (mg), larval period (days), pupal weight (mg) and pupal period (days) of *T. castaneum* reared on fresh food (control) and food treated with differnt doses of triflumuron*.

Dose (ppm)	Larval weight mean ± sd (mg) 10th day	16th day	Larval period mean ± sd (day)	Pupal weight mean ± sd (mg)	Pupal period mean ± sd (day)
Control	1.03 ± 0.04 (0.6 – 1.6)	3.23 ± 0.04 (2.7 – 3.8)	19.10 ± 0.07 (18 – 20)	2.93 ± 0.05 (2.3 – 3.4)	5.43 ± 0.13 (4 – 7)
2.5×10^{-7}	0.45 ± 0.03 (0.2 – 1.0)	1.76 ± 0.09 (1.0 – 2.9)	27.10 ± 0.85 (21 – 39)	2.32 ± 0.06 (1.8 – 3.0)	6.50 ± 0.19 (5 – 8)
2.5×10^{-8}	0.51 ± 0.04 (0.3 – 1.0)	1.76 ± 0.09 (1.7 – 3.5)	24.30 ± 1.20 (20 – 52)	2.37 ± 0.06 (2 – 8)	6.42 ± 0.09 (5 – 8)
2.5×10^{-9}	0.83 ± 0.3 (0.4 – 1.2)	2.81 ± 0.08 (1.7 – 3.5)	20.00 ± 0.31 (19 – 22)	2.68 ± 0.05 (2.0 – 3.4)	6.13 ± 0.06 (6 – 7)

NB * Mean of 30 larvae/pupae
Number in parentheses indicate minimum and maximum values

Table 2. Effect of triflumuron on the pupal formation, adult emergence, sex-ratio and adult popultation of *T castaneum*

Dose (ppm)	Pupal formation No of pupae formed(%)	Adult emergence No. of adults emerged(%)	Sex-ratio No. of male Female (ratio)	Adult population No. of adults	% reduction in adult progeny
Control	289(96.33)	289(96.33)	153 136 (1.12 1.00)	3735	–
2.5×10^{-7}	16(5.33)	11(3.67)	8 8 (1 1)	1505	59.71
2.5×10^{-8}	20(6.67)	16(5.33)	14 6 (2.33 1.00)	2570	31.19
2.5×10^{-9}	181(60.33)	123(41)	108 73 (1.48 1.00)	3105	16.87

NB No of larvae used in each treatment is 300

Table 3. Percentage of deformities produced in *T. castaneum* by triflumuron.

Dose(ppm)	No. of individual survived($n = 300$)	Percentage of deformity(no.) LPI	PAI	Adult	Total no. of deformity(%)
Control	289	–	–	–	–
2.5×10^{-7}	37	13.50(5)	16.21(6)	29.73(M4,F7)	22(7.33)
2.5×10^{-8}	61	29.51(18)	18.03(11)	19.67(M9,F3)	41(13.67)
2.5×10^{-9}	280	7.14(20)	5.36(15)	22.86(M49,F15)	99(33.00)

NB Total number of larvae used in each experiment, $N = 300$
LPI = larval-pupal intermediate form,
PAI = pupal-adult intermediate form,
M = male, F = female

Table 4. Morphological deformities in *T. castaneum* due to larval exposure to triflumurom

larval-pupal intermediate	Pupal-adult intermediate	Adult deformity
Pupal head with larval body	Adult with pupal skin	Small sized but perfect adult
Pupa with larval skin	Adult head with pupal abdomen	Small sized with crumpled wing
—	Small pupa unable to emerge as adult	Incomplete elytra
—	—	Wings spreaded widely
—	—	Small winged
—	—	Bent abdomen(humped back)

Table 5. Effect of triflumuron on the egg of *T. castaneum*

Exposure time	Dose(ppm)	Percentage of egg mortality at different ages of egg(hrs)			
		0 – 24	24 – 48	48 – 72	72 – 96
144hrs	2.5×10^{-7}	50.44	52.78	54.22	52.00
	2.5×10^{-8}	52.89	53.33	52.89	51.33
	2.5×10^{-9}	51.11	49.00	46.56	50.56
	Control	7.00			
24hrs	2.5×10^{-7}	57.22	54.56	55.56	56.11
	2.5×10^{-8}	56.22	53.78	55.44	52.44
	2.5×10^{-9}	56.56	51.56	53.78	53.00
	Control	7.00			
48hrs	2.5×10^{-7}	55.67	56.44	51.44	54.44
	2.5×10^{-8}	55.56	53.00	48.00	52.33
	2.5×10^{-9}	54.22	52.33	48.44	48.22
	Control	7.00			

The delayed effect of larval treatment also resulted in significant decrease in the percentage of pupation and adult emergence in *T. castaneum* (Table 2). Which is to some extent similar to the findings of the previous works with *T castaneum* (Ishaaya et al., 1981; Mian and Mulla, 1982a; Eisa et al., 1984, 1986; Ishaaya and Yablonskii, 1987), susceptibel and resistant strains of *T. castaneum* (Parween, 1996), *T. confusum* (El-Sayed et al., 1984 – 1985), *Oryzaephilus surinamensis* and *R. dominica* (Mian and Mulla, 1982a) and *T. granarium* (Saxena and Kumar, 1991, Soltani et al., 1993).

The sex-ratio of the beetles was observed to deviate from 1 1 in triflumuron treated beetles. At doses of 2.5×10^{-8} and 2.5×10^{-9} ppm the numbers of males was 1.5 and 2 times higher than the number of females, respectively. A similar result was observed in 0.05 ppm triflumuron treated *T. confusum*, where the number of males was three times higher than the females (El-Sayed et al., 1984-1985).

In treated food, the larval survival was decreased and deformed individuals were produced in larval, pupal and adult stages of the beetles. The morphological abnormalities were mainly due to the failure of ecdysis(Table 4). Partial ecdysis of the adults as shown by the abnormal elytron, was also observed among the treated individuals. Similar abnormalities of the elytra were reported from diflubenzuron treated *T. granarium* (Soltani et al., 1984) Morphogenetic abnormalities in stored product insect species due to BPU treatment were also reported by Fazardo and Morallo-Fejesus(1980), Saxena and Mathur(1981), Webley and Airey(1982), Eisa et al., (1984), Faragalla et al., (1985), Rup and Chopra(1987), Gazit et al., (1989), Saxena and Kumar (1991), Soltani et al., (1993) and Parween(1996).

The contact effect of triflumuron caused about 50% egg mortality in *T. castaneum*. Egg age or exposure period did not affect the ovicidal effect of the compound. Though BPUs are mainly stomach toxicants, they also show contact activity (Fox, 1990). Triflumuron caused 100% egg mortality in *O. surinamensis*, *R. dominica* and *T. castaneum* when they were exposed for four weeks (Mian and Mulla, 1982a). Similar results were reported by Carter (1975), Eisa et al. (1986), Saxena and Mathur(1981) and Saxena and Kumar (1989). Saxena and Mathur (1981) explained that BPU's action decreased the rigidity of the cuticle of the developing larva inside the egg and as a result they were unable to hatch. Low concentrations of different

BPU compounds were found effective to control the progeny of several stored product insect pests including the internal feeders of grains. When adults of *R dominica*, *T. castaneum*, *T. confusum* and *Sitophilus oryzae* were reared for three weeks on diflubenzuron treated grains, progeny production was completely inhibited for one year (Faragalla et al., 1985). A significant reduction in the progeny of *R. dominica* and *T. castaneum* was observed for 12 months as a result of two weeks parental exposure to 1 to 10 ppm of triflumuron (Mian and Mulla, 1982a). Hundred percent progeny control in *T. castaneum* by triflumuron was reported by Eisa et al. (1984) and Eisa and Ammar (1992) for nine months. Treatment of 10 ppm of triflumuron to *R. dominica*, *S. zeamais* and *Prostephanus truncatus* reduced more than 90% population over a period of 12 months (Dales et al , 1994). Control of adult population of both susceptible (FSS II) and resistant (CTC 12) strains of *T. castaneum* exposed to medium treated with triflumuron (Parweeen, 1996) and cyromazine (Mondal and Port, 1995). In the present study three months parental exposure to 2.5×10^{-7} ppm triflumuron caused 59.71% reduction of the adult progeny. Reduction of *T. castaneum* population by long exposure of the beetles to triflumuron probably resulted due to the cumulative effects of egg mortality and inhibition of larval development which ultimately inhibited the emergence of viable adults.

Conclusion

In the present experiment, both larval growth and adult population of *T. castaneum* were significantly reduced by triflumuron. Moreover, contact activity of the compound to the eggs caused 50% mortality of eggs of different ages. Triflumuron is known to be non-toxic to the human being and beneficial insects (Fox, 1990) and in the present investigation low doses of triflumuron were found to control the larval development of *T. castaneum*. Considering the present results it can be concluded that triflumuron may be a useful control agent against the stored product insect pests.

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