

Inhibitory Effect of Buprofezin on the Progeny of Rice Weevil, *Sitophilus oryzae* L. (Coleoptera: Curculionidae)

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Abstract

The rice weevil *Sitophilus oryzae* (L.) (Coleoptera: Curculionidae), is an important pest of stored grains throughout the world. Different biopesticides and synthetic insecticides have been used for a long time to minimize the rice weevil infestation but their efficacy is not satisfactory yet. The climate and storage conditions are highly favorable for quick progeny which may be responsible for lower efficiency of insecticides and higher level of infestation. Insect growth regulators (IGRs) like buprofezin are the semi-synthetic insecticides, mimic the insect-produced hormones and don't kill the insects directly but reduces pest populations by affecting mating behavior, reproduction, egg viability, pre and post embryonic development etc. Laboratory experiments were conducted from August to November 2012, to evaluate the efficacy of buprofezin on the mortality and the suppression of progeny production of rice weevil. Ten adult rice weevils were exposed on three types of rice grains (long, medium and short) treated with buprofezin at 100, 200 and 300 ppm. Mortality was counted at 15, 21 and 28 days after treatment while adult progeny was counted at 6, 7 and 8 weeks after buprofezin treatment to get a new generation. The data showed that buprofezin had no direct effect on the mortality of rice weevils regardless the concentrations. Buprofezin at 300 ppm in rice grains significantly inhibited progeny production while lower doses (200 and 100 ppm) had no significant effect but virtually reduces progeny number. Types of grains were not factor for increasing or decreasing the rice weevils populations. In conclusion, buprofezin caused decreasing progeny productivity by *S. oryzae* (L.) with increasing concentrations regardless the types of rice grains.

Keywords: *Sitophilus oryzae*; Buprofezin; Mortality; Progeny inhibition

Introduction

Stored grain pests are a great problem in Bangladesh and about 5-8% of the food grains, seeds and different stored products are lost annually due to stored pests infestation [1]. About 13 species of insects have been recorded from Bangladesh which loss about 15% of stored rice [2]. Among them, rice weevil, *Sitophilus Oryzae* (L.) (Coleoptera: Curculionidae) that is widely distributed, is one of the most destructive insect pests which cause severe economic loss. Rice weevils can cause losses to grain in storage, either directly through consumption of the grain or indirectly by producing 'hot spots' causing loss of moisture and thereby making grain more suitable for their pests [3]. Therefore, the effective control of this insect has long been the goals of entomologists [4]. Uncertain climatic condition, storage with debris, poor sanitation and inappropriate storage facilities encourage insect attack. In tropical countries like Bangladesh, the climate and storage conditions are highly favorable for insect growth and development [5].

Various synthetic insecticides with different mode of action are currently used to control rice weevil infestation in stored condition but their efficacy as well as safety is really questionable. Recently, different biopesticides have been used to control the rice weevil infestations [6-8]. Concern about the impact of pesticides on both health and environment has resulted in the search for alternative control measures for stored-product insect pests. Among such alternatives, insect growth regulators (IGRs), a class of biorational compounds that mimic insect-produced hormones and potentially reduces pest populations through endocrine disruption. Because of their selectivity these compounds appear to fit the requirements for third generation insecticides, environmentally benign and safer grain protectants. There are three groups of insect growth regulators e.g. juvenoids or anti-juvenoids, ecdysone inhibitor and chitin synthesis inhibitor (CSI). IGRs are potentially used to control insect pests either in the field or in stored condition. Like other conventional insecticides, IGRs don't kill the insect directly but

reduces pest populations by affecting mating behavior, reproduction, egg sterility, pre and post embryonic development etc. Methoprene and hydroxyphenoxypyrene are commonly used IGRs against stored-product pests [9-11]. In recent years, buprofezin became a dominant IGR in different Asian countries like China, Japan, and India to control the field pests effectively [12-14]. The exact mode of action of buprofezin in the insect body is not known yet but adult females that are sprayed directly or come into contact with wet residues, lay sterile eggs and down reproduction capability, ultimately reduces the number of individuals in the next generation [15]. In addition, buprofezin has been shown to have vapor activity, which means that insects not directly treated may still be killed [15]. There is limited information about the efficacy of buprofezin on the mortality as well as inhibition of progeny production by *S. oryzae* in stored condition.

Therefore the present study was conducted to elucidate the efficacy of buprofezin (a chitin synthesis inhibitor) on the adult mortality as well as inhibition of progeny production by *S. oryzae* (L.).

Materials and methods

Culturing of *S. oryzae*

The initial adult populations of *S. oryzae* were obtained from the

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Received November 16, 2013; Accepted November 13, 2013; Published December 17, 2013

Citation: Das G (2013) Inhibitory Effect of Buprofezin on the Progeny of Rice Weevil, *Sitophilus oryzae* L. (Coleoptera: Curculionidae). J Biofertil Biopestici 4: 140. doi: [10.4172/2155-6202.1000140](http://dx.doi.org/10.4172/2155-6202.1000140)

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stock culture of the laboratory of entomology, BAU, Mymensingh. For the continuous supply of experimental insects, adult rice weevils were cultured in the same laboratory at 60-70% relative humidity and 28-32°C temperature. Before culturing, all equipments and glassware's were disinfected with 70% ethyl alcohol to protect stock culture from microbial infection and safeties of the insects. Adults of *S. oryzae* were released into 200g of disinfected rice grains in plastic containers @25 numbers/container of mixed age and sex. Mouth of the container was capped with muslin cloth fastened with rubber band to ensure proper ventilation as well as prevent the escape and was incubated for 15 days. On the 15th day, the released adults were removed by sieving and the rice grains were kept undisturbed for 2 months for emergence of fresh adults. Emerged adults were collected daily to maintain the same age and released in separate containers for continuous mass culturing. Adults of 5-7 days old age were used for the experimentation.

IGR and doses

Three doses of buprofezin 100, 200 and 300 ppm will be tested and accordingly concentrations were made using distilled water.

Bioassay

Three types of rice grains were used in this study as follows; long, medium and short grain. From each rice type, three 40 g subsamples were weighed and each 40 g was sequentially treated with three concentrations 100, 200 and 300 ppm using micro-sprayer (1 ml/40 g rice) and air dried for 10 minutes. Therefore, a total of 9 treatments will be made for 3 rice types using 3 concentrations. Three replications were maintained for each concentration and arranged following Completely Randomized Design (CRD). Each rice types were also treated with distilled water those were considered as negative control. Then 40 g of buprofezin-treated or water-treated grains were confined with 10 freshly emerged adults in 150 cm diameter petridishes for 10 days for oviposition and kept them undisturbed.

Data collection

The adult mortality was assessed from each treatment combination at 14, 21 and 28 days after buprofezin treatment. After 28-day mortality observation, dead and live adults were removed by sieving from each petridishes and commodity was left as the previous conditions. 40 days

after their confinement in petridishes (or 40 days after treatment), number of F1 adults present in each treatment was counted to record the progeny build up and counting was continued for an additional period of 20 days. No data were collected on egg, larva or pupae either from treated or control rice grain.

Data analysis

All observations were corrected by using the Abbott's formula [16]. Data obtained were subjected to analysis of variance (ANOVA) and means were separated by Fishers protected least significant difference (LSD) test. Values were represented as mean \pm SEM.

Results

Effect of buprofezin on the mortality of adult *S. oryzae*

Buprofezin-treated as well as water-treated (control) mortality of *S. oryzae* has shown in Table 1. Data clearly showed that buprofezin had no significant effect on the mortality of adult rice weevil in comparison with that in the control. Compared to the control (long grain, 4.53 \pm 1.0; medium grain, 3.43 \pm 1.0; short grain, 3.97 \pm 0.6) the mean mortality level was gradually increased with the increase of concentration while the highest mortality was recorded from 300 ppm for all grain types (long grain, 6.06 \pm 1.3; medium grain, 6.20 \pm 0.3; short grain, 6.37 \pm 0.6) but the differences were not significant between control and buprofezin-treated rice grains. On the other hand, mortality level was not increased with the increase of time for all types of rice grains. It was noted that the larval and pupal mortality was not recorded in the present study.

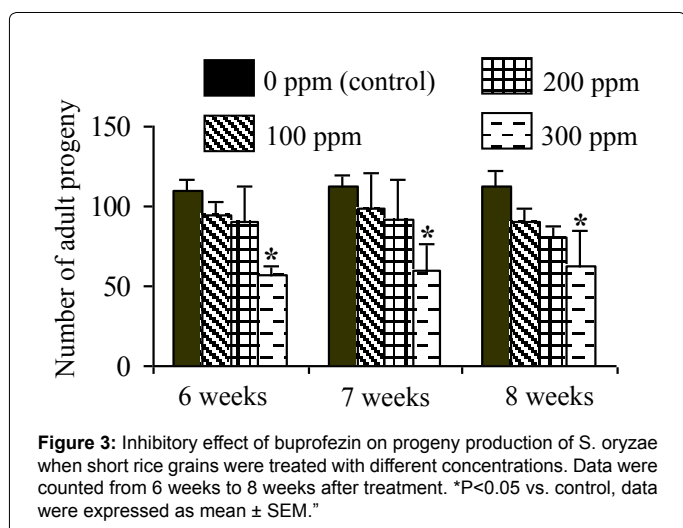
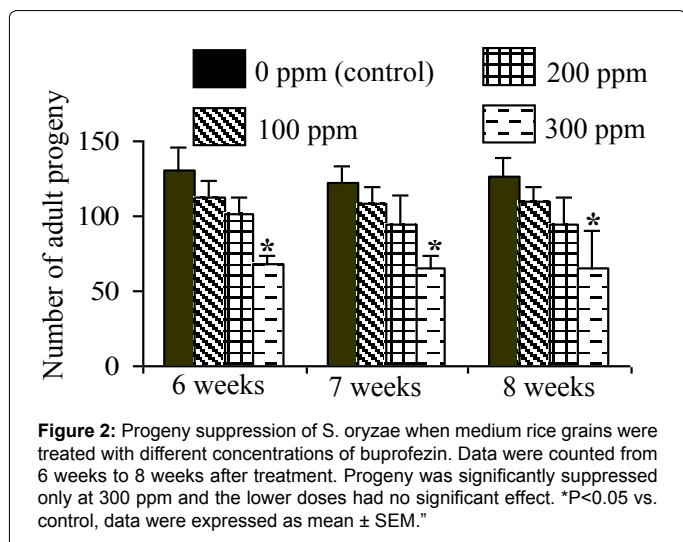
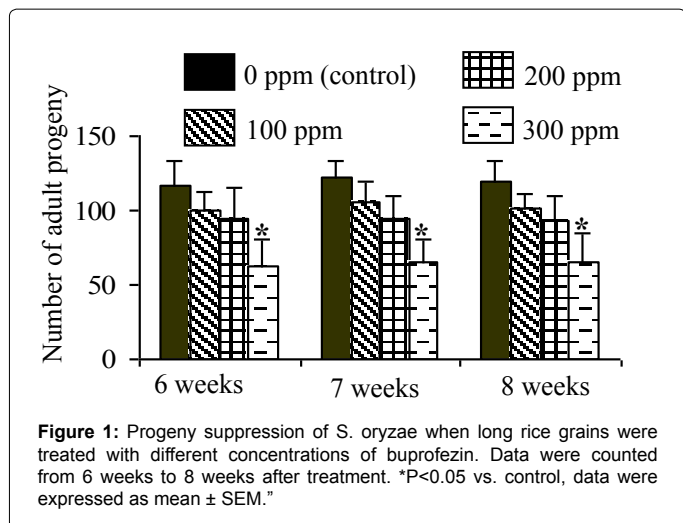
Effect of grain types on the buprofezin-induced mortality of *S. oryzae*

It was assumed that progeny production by *S. oryzae* in different types of rice grains may be influenced by the action of buprofezin. To confirm this hypothesis, three types of rice grains (e.g. long, medium and short) have been selected in this study to elucidate which grain type is suitable for buprofezin action and hence buprofezin-treated mortality is also increased. There has no significant difference among the rice grain types based on the mortality (Table 1). In all types of rice grains, the mortality level was little higher at the highest concentration (300 ppm) which was followed by 200 and 100 ppm, respectively in comparison with that in the water-treated grains. But the differences

Table 1: Mortality (%) of adult *S. oryzae* when exposed on three rice types treated with 100, 200 and 300 ppm of buprofezin.

Rice grain types	Concentrations (ppm)	Mortality (%) at different days after buprofezin treatment			
		14 days	21 days	28 days	Mean
Long grain	0 (Control)	4.0 \pm 0.3	4.4 \pm 0.8	5.2 \pm 1.9	4.53 \pm 1.0
	100	3.8 \pm 1.5	3.0 \pm 0.4	4.5 \pm 0.2	3.77 \pm 0.7
	200	4.1 \pm 0.9	3.5 \pm 0.5	4.6 \pm 0.3	4.06 \pm 0.6
	300	6.1 \pm 1.1	5.8 \pm 1.6	6.3 \pm 1.2	6.06 \pm 1.3
Significant level		NS	NS	NS	NS
Medium grain	0 (Control)	3.9 \pm 1.2	4.1 \pm 1.5	2.3 \pm 0.3	3.43 \pm 1.0
	100	5.1 \pm 0.9	4.5 \pm 1.1	4.8 \pm 0.6	4.80 \pm 0.9
	200	5.2 \pm 0.8	5.9 \pm 0.5	5.3 \pm 0.9	5.47 \pm 0.7
	300	6.5 \pm 0.2	6.2 \pm 0.3	5.9 \pm 0.5	6.20 \pm 0.3
Significant level		NS	NS	NS	NS
Short grain	0 (Control)	4.9 \pm 0.5	3.0 \pm 0.2	4.0 \pm 1.1	3.97 \pm 0.6
	100	5.0 \pm 0.5	4.9 \pm 0.7	4.6 \pm 0.5	4.83 \pm 0.6
	200	6.2 \pm 0.5	4.8 \pm 1.5	4.9 \pm 1.5	5.30 \pm 1.2
	300	5.9 \pm 0.5	6.8 \pm 0.2	6.4 \pm 1.0	6.37 \pm 0.6
Significant level		NS	NS	NS	NS

Values are represented here as Mean \pm SEM. Buprofezin (any of the concentration) had no significant effect on the mortality of adult *S. oryzae* versus control or water-treated rice grain. Types of rice grain also had no effect on the mortality of adult *S. oryzae*. NS=Not significant.



were quite insignificant between controls (water-treated) as well as buprofezin-treated rice grains with different concentrations from 100 to 300 ppm. It is possible that more than 300 ppm concentration will

be able to suppress rice weevils progeny significantly although this dose has not been tested in the current study.

The same number of adult rice weevils was carefully tested for all the buprofezin doses to get an accurate mortality level for all the treatments.

Adult progeny production by *S. oryzae* in buprofezin-treated long rice grain

As mentioned in the methodology, 10 pairs of adults weevils were released in buprofezin-treated (100, 200 and 300 ppm) long rice grain to observe whether progeny production is inhibited by different concentrations of buprofezin. Progeny production was also observed in water-treated rice grains to confirm the buprofezin effect on the progeny production by *S. oryzae*. It is noted that the larval and pupal mortality was not recorded in the present study. Adult progeny were counted at 6, 7 and 8 weeks after adults released in buprofezin-treated rice grains. Progeny production of *S. oryzae* was decreased with increasing buprofezin concentrations (Figure 1). Progeny production was reduced by both 100 and 200 ppm of buprofezin but the difference was insignificant when compared with water-treated control. On the other hand, mean number of adult progeny was significantly ($p < 0.05$) suppressed when rice grains were treated with 300 ppm of buprofezin in comparison with that in the water-treated control regardless the time after treatment. Moreover, the level of progeny suppression differed insignificantly when compared among the concentrations.

Adult progeny production by *S. oryzae* in buprofezin-treated medium rice grain

Adult progeny were counted at 7, 8 and 9 weeks after adults released in buprofezin-treated rice grains. Like as long grain, progeny production was significantly inhibited ($p < 0.05$) by buprofezin when rice grains were treated with highest concentration (300 ppm) (Figure 2) regardless the time. On the other hand, lower doses (100 and 200 ppm) had no significant effect on the suppression of progeny production by *S. oryzae* (Figure 2) although number was reduced virtually compared to the control.

Adult progeny production by *S. oryzae* in buprofezin-treated short rice grain

The *S. oryzae* progeny was significantly inhibited ($p < 0.05$) by buprofezin only when rice grains were treated with 300 ppm and the lower doses (100 and 200 ppm) had no significant effect on the suppression of progeny production by *S. oryzae* (Figure 3) regardless the length of time.

Progeny production versus types of rice grains

Three types of rice grains were used in this study to know the better action of buprofezin as well as tendency of progeny production among the rice grain types. Number of progeny production by *S. oryzae* was not affected by morphological differences of rice grain. The results also showed that the tendency of production of progeny by adult rice weevil was similar to the length of time. A control group (water-treated) was included in the experiment to make a comparison with buprofezin-treated group.

Discussion

The post-harvest losses of rice grains by rice weevil, *Sitophilus oryzae* are significant. Treatment with chemical insecticide has great health hazard as well as environmental contaminations. Insect growth regulators are reported to environmentally safe, biodegradable and

non-toxic to human and they are considered as third generation insecticides. Various insect growth regulators are potentially using to control the field pests while their uses are mostly limited against stored-pests [12-14]. There are three groups of IGRs; juvenile hormone analogue, ecdysteroid receptors agonists or antagonists and chitin synthesis inhibitors. IGRs are directly not toxic to insects but potentially reduce pest populations by disrupting normal growth and development through multiple targets like reduction of egg viability and hatchability, affecting pre or post embryonic development, abnormal larvae or pupae formation as well as disruption of chitin synthesis [17,18]. Buprofezin has multiple targets to reduce pest populations but the main target is disruption of chitin synthesis [17-20]. Buprofezin is potentially used in the field condition to control lepidoptera, diptera or coleopteran pests [12,21-23] while against stored-pests are not well reported.

The current study was conducted to elucidate the efficacy of buprofezin on the mortality and inhibition of progeny production on different types of rice grains. The major finding of this study is that buprofezin has no direct effect on the mortality of adult *S. oryzae* but it significantly inhibited progeny production. It was interesting that progeny production was inhibited significantly ($P < 0.05$) only at 300 ppm of buprofezin while lower doses (100 and 200 ppm) had no significant effect although number of progenies were virtually reduced either by 100 or 200 ppm. The mechanism of progeny inhibition by buprofezin is not clear yet but it has been reported that adult females that are sprayed directly or come into contact with wet residues of buprofezin, lay sterile eggs or down reproduction capability, ultimately reduces the number of individuals in the next generation [15]. It was not also clear yet why progeny was potently suppressed only at 300 ppm but not at 200 or 100 ppm, the possible cause may be penetration level. Maximum penetration of buprofezin molecules might be occurred at 300 ppm by vaporization because buprofezin has been shown to have vapor activity [15]. Either mortality or progeny suppression was not affected by morphological features of rice grains which also raises the possibility that rice hull for each type was similar thickened or with similar features. Arthur [11] also stated that progeny production of *Rhizopertha dominica* was not affected by rice grain types when adult *R. dominica* was exposed on methoprene-treated rice grains.

In Bangladesh, rice weevils take about 35 to 40 days to reach in adult after oviposition although it depends on the prevailing temperature. Therefore, the first progeny production was counted at 6 weeks after treatment and counting was continued for an additional period of 2 weeks to assess whether progeny production really changed or not over time. Data clearly showed that mean number of progeny production was almost stable for 6, 7 or 8 weeks after treatment which raises the possibility that adults weevils were laid eggs almost in the same time after exposure on the treated rice grains. It is noted that culturing of *S. oryzae* were done carefully so that almost same aged adults can be released onto treated rice grains. IGRs like methoprene, hydroxypropryl, pyriproxyfen are widely using against rice weevils and other stored pests but the use of buprofezin against stored-pests especially rice weevils is not well reported. Despite the demonstrated effectiveness of some IGRs, their practical application presents certain problem against rice weevils because this insect spend a large part of their life cycle inside the grains kernel and the insects are surrounded by food, are not easily accessible to control insects. Current study clearly showed that buprofezin was not effective to kill adult weevils even with higher concentration and this might be caused due to little contact of insects with buprofezin. In contrary, buprofezin significantly inhibited progeny build up at 300 ppm but not at 100 or 200 ppm. As reported previously, buprofezin has volatile properties, and therefore, the highest

concentration of buprofezin was able to contact with insects and inhibited progeny production possibly through reduction of mating, egg viability, hatchability as well as affecting the pre and post embryonic development.

IGRs have several advantages over neurotoxic insecticides. They affect development at an early stage and, therefore, can slow down population build up, particularly where there is no constant pressure of re-infestation. They can be used in minute amounts and pose no residues problems. They are reported to be non-toxic to mammals and produce no teratogenic or mutagenic effects in warm-blooded animals even with high concentrations [24]. Therefore, uses of IGRs especially buprofezin may be the potential alternatives of various toxic insecticides against rice weevils infestation in Bangladesh. However, it needs further investigations to explore the molecular mechanisms of suppression of progeny build up by *S. oryzae* using buprofezin.

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Citation: Das G (2013) Inhibitory Effect of Buprofezin on the Progeny of Rice Weevil, *Sitophilus oryzae* L. (Coleoptera: Curculionidae). J Biofertil Biopestici 4: 140. doi: [10.4172/2155-6202.1000140](https://doi.org/10.4172/2155-6202.1000140)

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