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Laboratory evaluation of Buprofezin on the mortality of Brown planthopper, *Nilaparvata lugens* (Delphacidae: Hemiptera)

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Abstract
Buprofezin (Award 40 SC), a chitin synthesis inhibitor (CSI) has been evaluated on the mortality of brown planthopper in the laboratory condition of Department of Entomology, Bangladesh Agricultural University, during the period from April to August, 2014. The mortality was observed using three concentrations viz. 100, 200 and 300 ppm with different application methods like topical, leaf-dip and combination method. The mortality data was observed at 1, 3, 5 and 7 days after treatment (DAT) application. Buprofezin was found to be highly effective against BPH and the mortality was clearly dose and method dependent. The highest mortality (90.55%) was recorded from 300 ppm which was followed by 200 (83.06%) and 100 (43.47%) ppm of Buprofezin respectively. The dose, 100 ppm was found comparatively less effective than rest of the two doses. Among application methods, maximum mortality (90.55%) was found from combination method i.e. when both BPH and rice plants were treated with different concentration of Award 40SC which was followed by leaf-dip (87.32%) and topical (76.70%) application methods respectively. For all application methods, the mortality was found to be insignificant compared to control at 1 DAT but increased significantly at 2 DAT and reached to the highest level at 5 DAT which was persisted at least up to day 7.

Keywords: Buprofezin, Laboratory, Mortality, *Nilaparvata lugens*

1. Introduction

The brown planthopper, *Nilaparvata lugens* (Stål) (Homoptera: Delphacidae), is an economically important insect on rice in Asia [1]. It has become a serious threat to rice production throughout Asia. The losses to rice production in Asia caused by *N. lugens* have been estimated as more than 300 million dollars annually. This monophagous pest causes severe damage to rice plants through direct sucking, ovipositing and virus disease transmission. Because of its highly adaptive capacity to changing cultural practices and high reproductive potential, frequent chemical treatments to every generation are necessary to bring the insect populations under control [2, 3]. The control of *N. lugens* has primarily relied on various types of insecticides from different groups like organophosphorous, organocarbamate, pyrethroid, nicotinoids etc. [4].

In the search for safer insecticides technologies, i.e. more selective mode of action and reduced risks for non-target organisms and the environment, progress has been made in the last 20 years with development of natural and synthetic compounds capable of interfering with the processes of growth, development and metamorphosis of the target insects. These chemicals have been called insect growth regulators (IGRs) or third generation insecticides. IGRs differ widely from the commonly used insecticides as they exert their insecticidal effects through their influence on development, metamorphosis and reproduction of the target insects by disrupting the normal activity of the endocrine system.

Buprofezin is an insect growth regulator (IGR) especially chitin synthesis inhibitor (CSI) that disrupts the development of immature forms by interference with chitin synthesis. This is highly effective against the sucking insects like planthoppers, leafhoppers, whiteflies, mealy bugs etc. [5, 6, 7]. It was also found to be effective against various Lepidopteran larvae, spiders, mites etc. [7-13]. It was reported that buprofezin is an eco-friendly biorational pesticide that is safe for non-target organisms, highly biodegradable and action is target specific [14]. Till now, only a few works have been conducted on the efficacy of Buprofezin against planthoppers or leafhoppers both in the field and laboratory condition. The present investigation was, therefore, planned to evaluate the efficacy of Buprofezin (Award 40 SC) against brown planthopper through different application methods under laboratory conditions.

2. Materials and Methods

The study was conducted in the laboratory of Department of Entomology, Bangladesh Agricultural University, during the period from April to August, 2014.

2.1 Mass rearing of *N. lugens*

For a continuous supply of nymphs and adult BPH, mass rearing was done in the laboratory. BPH was reared in the controlled temperature room (26-27 ^{0}C) to build up large populations. They were reared in the Mylar cage. The BPH populations and TN-1 (Taichung Native-1), rice variety of 15-20 days old seedlings were collected from field laboratory. Immediately after collection of gravid female BPH through aspirator, the insects were placed in the Mylar cage where the TN-1 rice plants were previously grown in the earthen pot. The opening of the Mylar cage was then closed with pieces of fine nets and fastened with rubber bands. In this way, gravid female BPH were released into other Mylar cage. Cages were regularly monitored for egg laying, hatching, growth and development of nymphs and supplement of fresh rice plants for feeding. Usually 3rd and 4th instar nymphs were used for insecticidal test. Gravid females were not used for insecticidal test.

2.2 Specifications of treatments

Experiments consisted of three treatment combinations. Three doses of Buprofezin (Award 40 SC) viz. 100, 200 and 300 ppm were provided as treatments. Each treatment was replicated thrice and 20 nymphs were used for each replication.

2.3 Methods of treatment application

Treatments were applied through three different methods viz. topical, leaf-dip and combination.

2.3.1 Topical application method: In this method, BPH populations were directly treated (using micro-sprayer) with different concentrations of Award 40 SC (Buprofezin). Treated BPH were then transferred on untreated rice plants using fine brush and covered with Mylar-cage.

2.3.2 Leaf-dip method: In this method, rice plants were dipped in Award 40 SC solutions for 5-10 seconds and rice plants were then dried. After that rice plants were covered with Mylar-cage and untreated BPH populations were then transferred on the treated rice plants using fine brush.

2.3.3 Combination (topical + leaf-dip) method: Both BPH and rice plants were treated with different concentrations of Award 40 SC. After that rice plants were dried for a while. Then Award 40 SC-treated BPH populations were transferred on Award 40 SC-treated rice plants using fine brush and covered with Mylar-cage.

2.4 Data collection: Data on the mortality of BPH was observed at 1, 3, 5 and 7 days after treatment (DAT) application. The mean percentage of BPH mortality was calculated using the following formula:

$$\textbf{\% Mortality} = \frac{\textbf{Po}}{\textbf{Pc}} \textbf{ X 100}$$

Where,
Po = Number of BPH died due to treatment application
Pc = Total number of treated/untreated BPH

2.5 Statistical analysis

The recorded data were compiled and tabulated for statistical analysis. Analysis of variance (ANOVA) was done with the help of computer package MSTAT. The mean differences among the treatments were adjudged with Duncan's Multiple Range Test (DMRT) and Least Significant Difference (LSD).

3 Results

3.1 Efficacy of Award 40 SC (Buprofezin) on the mortality of *N. lugens* through topical application method

The mortality of BPH was significantly increased when brown planthoppers were directly treated with different concentrations of Award 40 SC (P<0.01; Table 1). The initiation of action of Award 40 SC was found to be slow i.e. no significant level of mortality was observed up to 48 h after treatment application which clearly indicates that Award 40 SC has no acute action. The significant level of mortality was found at 3 days after treatment (DAT) application for all doses and the mortality reached at the highest level at 5 DAT which has persisted at least up to day 7. At 5 DAT, the highest mortality was obtained from the dose 300 ppm (76.70%) which was followed by 200 ppm (54.10%) and 100 ppm (39.70%) respectively (Table 1). At 7 DAT, the mortality level was increased slightly compared to 5 DAT and there had insignificant differences between 5 and 7 DAT based on the mortality.

Table 1: Efficacy of different doses of Award 40 SC on the mortality of *N. lugens* when applied topically.

Treatments	Mean percent mortality of *N. lugens*			
	1 DAT	3 DAT	5 DAT	7 DAT
Award 40 SC @ 100 ppm	1.19	18.10c	39.70c	40.80c
Award 40 SC @ 200 ppm	2.90	22.40b	54.10b	61.12b
Award 40 SC @ 300 ppm	2.71	26.30a	76.70a	80.70a
Untreated control	1.56	4.78d	4.79d	4.79d
P-level	NS	0.01	0.01	0.01
CV%	5.43	5.28	2.55	3.85
SE (±)	0.42	4.68	15.08	16.21

In a column, means of similar letter (s) do not differ significantly as per DMRT. [DAT = Days After Treatment, NS = Not significant, P-level = Probability Level, CV = Co-efficient of Variation, SE = Standard Error]

3.2 Efficacy of Award 40 SC (Buprofezin) on the mortality of *N. lugens* through leaf-dip method

The mortality of BPH was significantly increased compared to water-treated control through leaf-dip method (P<0.01; Table 2). In this method, rice plants were dipped in different concentrations of Award 40 SC for 5- 10 seconds then dried the rice plants and then untreated BPH were released on treated rice plants. This was done to know whether Award 40 SC (Buprofezin) has any systemic action or not. Like as topical method, no mortality was observed up to 48 h after treatment application which further confirmed that Award 40 SC has no any acute action. The mortality level was increased gradually which has reached at significant level by day 3 compared to that of control. The highest mortality was observed at 5 DAT which has persisted at least up to day 7. At 5 DAT, about 87.32% mortality was recorded when rice plants were treated with 300 ppm of Award 40 SC which was followed by 200 (81.19%) and 100 ppm (40.80%) respectively. The mortality was insignificantly increased at day 7 in comparison with that of 5 DAT. The result clearly indicates that the lower dose (100 ppm) was not much effective to control BPH while 200 and 300 ppm were found very effective where more than 80% BPH were found to be died.

Table 2: Efficacy of different doses of Award 40 SC on the mortality of *N. lugens* through leaf-dip method.

Treatments	Mean percent mortality of *N. lugens*			
	1 DAT	**3 DAT**	**5 DAT**	**7 DAT**
Award 40 SC @ 100 ppm	1.19	20.47b	40.80c	46.14c
Award 40 SC @ 200 ppm	2.74	33.33a	81.19b	83.22b
Award 40 SC @ 300 ppm	3.19	33.18a	87.32a	88.32a
Untreated control	1.56	4.78c	4.79d	4.79d
P-level	NS	0.01	0.01	0.01
CV%	8.31	4.54	2.91	4.02
SE (±)	0.47	6.76	19.25	19.37

In a column, means of similar letter (s) do not differ significantly as per DMRT. [DAT = Days After Treatment, NS = Not significant, P-level = Probability Level, CV = Co-efficient of Variation, SE = Standard Error]

3.3 Efficacy of Award 40 SC (Buprofezin) on the mortality of *N. lugens* through combination (topical + leaf-dip) method

The highest mortality was found when rice plants and BPH both were treated with different concentrations of Award 40 SC compared to the individual application method or leaf-dip method (Table 3). No significant mortality was observed from 1 and 2 DAT which further confirmed that Buprofezin has no any acute action on the mortality of BPH. At 3 DAT, the mortality level increased significantly in comparison with that of control which reached at the peak level by day 5 and persists up to at least day 7. Approximately 90% BPH were died at day 5 when rice plants and BPH both were treated with 300 ppm of Award 40 SC which was followed by 200 ppm (83.06%) and 100 ppm (43.47%) respectively. The mortality was persisted up to day 7 and there had no significant differences between day 7 and 5 which raises the possibility that maximum mortality of BPH can be achieved within 5 days of treatment application. The lowest mortality was recorded from untreated control.

Table 3: Efficacy of different doses of Award 40 SC on the mortality of *N. lugens* through combination method.

Treatments	Mean percent mortality of *N. lugens*			
	1 DAT	**3 DAT**	**5 DAT**	**7 DAT**
Award 40 SC @ 100 ppm	3.86b	24.47b	43.47c	46.14c
Award 40 SC @ 200 ppm	4.18ab	30.81a	83.06b	84.64b
Award 40 SC @ 300 ppm	4.99a	33.33a	90.55a	90.55a
Untreated control	2.34c	4.78c	4.79d	4.79d
P-level	NS	0.01	0.01	0.01
CV%	13.94	6.85	3.74	3.75
SE (±)	0.55	6.46	19.80	19.86

In a column, means of similar letter (s) do not differ significantly as per DMRT. [DAT = Days After Treatment, NS = Not significant, P-level = Probability Level, CV = Co-efficient of Variation, SE = Standard Error.]

4. Discussion

In the present study, Buprofezin was evaluated with three doses viz. 100, 200 and 300 ppm with different application methods to confirm the efficacy as well as to know the route of entry (i.e. has any systemic action or not). For all treatment methods, the highest mortality of BPH was recorded at 5 DAT from 300 ppm (topical: 76.70%, leaf-dip: 87.32%, combination: 90.55% hill^{-1}) which was followed by 200 ppm (topical: 54.10%, leaf-dip: 81.19%, combination: 83.06%) and 100 ppm (topical: 39.70%, leaf-dip: 40.80%, combination: 43.47%) respectively. These results clearly indicated that the combination method was the most effective regarding the BPH mortality which was closely followed by leaf-dip method. Topical method has showed the least efficacy but moderately effective when applied only at higher doses (300 ppm). The lowest dose (100 ppm) was not much effective to control BPH. Moreover, Buprofezin showed comparatively slower action as no significant mortality was found within 48 hours of treatment application but increased significantly by day 3, reached to the peak level by day 5, and persisted at least up to day 7. Therefore, it has been confirmed that at least 3 days required getting significant level of mortality from IGRs (especially chitin synthesis inhibitor) which has absolutely fitted with IGRs mode of action. This finding is in close agreement with previous finding that Buprofezin 25 SC @ 200 g a.i./ha was more effective after 3 days up to the 14 days in reducing BPH, followed by Thiamethoxam 25 WG @ 25 g a.i./ha and Imidacloprid 200 SL @ 25 g a.i./ha. [15]. The present results may be supported by Uchida *et al.* where they have reported that the mortality of nymphs began to die after 84 hours of application of Buprofezin @ 50 ppm [16].

On the other hand, Buprofezin has both systemic and contact action although systemic action was more potent than contact action. About 75% of the BPH populations were died through topical application method but this mortality level reached to 90% through leaf-dip method with same concentrations (300 ppm). These findings raises the possibility that Buprofezin works more effectively in the endocrine-pathway of BPH than its cuticular-pathway to inhibit chitin bio-synthesis [9]. Moreover, the action of Buprofezin was found to be slow as no mortality was found within 48 hrs of treatment application but significantly increased at 3 DAT which was peaked by 5 DAT. Buprofezin does not works in the central nervous system, rather it kills insects by preventing moulting through the inhibition of chitin bio-synthesis. For that reason, Buprofezin works slowly compared to other neurotoxic insecticides [13].

5. Conclusion

From the present investigation it has been concluded that Buprofezin (Award 40 SC) was found to be very effective against *N. lugens*. It was also confirmed that Buprofezin has both contact and translaminar movement activity regarding the results of leaf-dip bioassay. Therefore, it could be used as the substitute to broad-spectrum neurotoxic insecticides for integrated pest management of *N. lugens*. Field experiments are needed to verify the present laboratory findings.

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