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Insecticide Hormoligosis on Brown Planthopper, *Nilaparvata lugens* (Stål) in Resistant and Susceptible Rice Varieties of Bangladesh

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Abstract: Studies were conducted to find the effect of hormoligant (sub-lethal doses of insecticides) on brown planthopper *Nilaparvata lugens* (Stål). The LC_{50} value of Ripcord 10EC both for 3rd and 5th instar nymphs of BPH was 0.0000142%. The LC_{50} value of Dimecron 100WSC and Diazinon 60EC for 5th instar nymphs of BPH were 0.00001987 and 0.000651%. But the effect of hormoligant (i.e. the effect of LC_{50} and LC_{10} dosages of Diazinon 60EC on the test populations) was observed in 1st, 2nd, 5th, and 6th generations of the treated populations; i.e. the populations in LC_{10} in BPH resistant (BRRI dhan 31) and susceptible varieties (BR 3) increased at the above generations compared to LC_{50} populations of BPH.

Key words: Hormoligosis, brown planthopper, *Nilaparvata lugens* (Stål), LC_{50}, insecticides

Introduction

The brown planthopper (BPH), *Nilaparvata lugens* (Stål), often called the pest of green revolution in tropical Asia is the potential pest of rice (Alam and Karim, 1986). Brown planthopper possesses a high reproduction potential. Under favourable conditions, as observed in the intensive rice cultivation areas of Bangladesh, a single pair of BPH can give rise to a population of about 400 millions in 85 days enough to cause complete damage to a rice crop of more than 3 hectare (Alam and Karim, 1986). Sub-lethal doses of insecticides used pose threats of resurgence of some insect pests and development of resistance against insecticides (Islam *et al.*, 2001).

In combating BPH, chemical control continues to play a major role in South-East Asian countries (Krishnaiah and Kalode, 1987). However, increased population levels of the pest following insecticides applications termed as "resurgence" was first reported from IRRI, Philippines (Anonymous, 1968). Later this phenomenon was found to be a common feature in Philippines (Chelliah, 1980), Solomon Islands (Stapley *et al.*, 1979), India (Anonymous, 1978), Indonesia (Oka, 1978) and Bangladesh (Alam and Karim, 1986).

The factors contributing to the resurgence of *Nilaparvata lugens* on rice in several Asian countries are the suppression of natural enemies following intensive broad-spectrum insecticide application; insecticide-induced plant growth; the increased feeding rate of *N. lugens* at sub-lethal doses of some resurgence-inducing insecticides; stimulation of reproduction (i.e. Hormoligosis) by the pest following insecticide application; changes in the nutrient contents of the plant following insecticide application; the sub-lethal doses, rates, timing and number of applications; methods of application; effects of insecticides on nymphs and adults; and genetic resistance of rice varieties (Chelliah and Heinrichs, 1984; Krishnaiah and Kalode, 1987). When certain insecticides are applied to rice varieties susceptible to the brown planthopper [*Nilaparvata lugens* (Stål)], a resurgence of the brown planthopper populations commonly occurs. The biochemical changes after insecticide treatment were not so distinctively observed in the resistant variety IR-36 as they were in the susceptible variety TN-1 (Buenaflor *et al.*, 1981).

Resurgence of insect pests and mites, and secondary pest outbreaks are commonly observed following pesticide applications. Decreased natural enemy populations are the major factors responsible for these phenomena, but insect or mite hormesis is a second factor. A major impact of hormesis is that it often leads to the need for repeated pesticide treatments and can result in a spiralling increase in the use of pesticides (pesticide syndrome) (Morse, 1998). Resurgence of the delphacid was induced by organophosphorus compounds, carbamates and synthetic pyrethroids, especially when applied at sublethal doses (Chelliah & Uthamasamy, 1986). The present studies have been taken to evaluate the effect of hormoligant (sub-lethal dosages of insecticides) on brown planthopper and also susceptibility test of BPH resistant and susceptible rice varieties by the hormoligant treated BPH populations.

Materials and Methods

The studies were conducted in laboratory and the greenhouse of the Bangladesh Rice Research Institute (BRRI) from April 1995 to December 1996.

Mass culture of BPH: To obtain BPH adults for the experiments the insect was mass reared in the greenhouse at about 32°C. Four to six weeks old potted BR3 plants were used for this purpose. The plants were cleaned and the outer leaf sheaths of the potted plants were removed to destroy the eggs of other insects if any and placed in iron framed (150 x 66 x 76 cm^3) rearing cages covered with fine mesh wire net. Gravid BPH adults were released on the potted plants for oviposition and they were kept there for 1 to 2 days and then the oviposited plants were shifted each day (depending upon the intensity of egg laying of BPH on the oviposited plants by visual observation, otherwise the oviposited plants will be damaged due to severity of egg laying and the population of BPH will decline and this is why it was checked everyday) to a series of such cages for hatching of the eggs. This was come to ensure supply of the insects of about the same age. Regular observation was done to keep the culture free from predators (specially green mirid bug). After hatching, the host plants were changed at 3-4 days interval to provide sufficient food for the development of the nymphs to adulthood. This cycle was maintained the required number of BPH adults and nymphs as and when needed for the experiment.

Hormoligosis studies of some commercial insecticides on BPH bioassay study-seedling dipping method: All treatment dosage dilutions were made with acetone. The stock solutions were prepared by dissolving the test insecticides separately in acetone. These were 0.000125% for Ripcord 10EC, 0.003125% for Dimecron 100WSC, and 0.001875% for Diazinon 60EC. Preliminary range tests were conducted with a number of concentrations before final test. Seven or eight concentrations were used in final bioassay. The serial dilutions were made using a factor of half (½) from higher to lower level.

Experimental procedure: Forty to forty-five days old BR3 seedlings were treated individually with different concentration of each insecticide by dipping the seedlings for 15 second. After dipping the treated tillers (seedlings) were kept on aluminum foil to dry up for 15-20 minutes under shade and then placed in the 70 ml capacity test tube with the help of forceps and were kept moist by placing wet cotton in the bottom of the tube. In Aus, twenty of 3rd and 5th instar nymphs of BPH (taken from mass culture populations) were released in Ripcord 10EC treated (different concentrations of insecticide) seedlings in each test tube and kept

in contact with the treated surface for 24 hours. While in T. Aman 10 of 5th instar nymphs of BPH were released in Dimecron 100WSC and Diazinon 60EC treated seedlings in different trials respectively. Mortality counts were taken 24 hours after insect release. After that the mortality counts were taken from every treatments, moribund insects were assumed to be dead.

Mortality in all treatments is corrected by Abbott's formula (Abbott, 1925).

$$\text{Corrected \% Mortality} = \frac{\text{\% Mortality in treatment - \% Mortality in control}}{100 - \text{\% Mortality in control}} \times 100$$

There were ten replications. Tillers (seedlings) treated with acetone alone served as the control. The LC_{50} values of the tested insecticides were calculated by analyzing the data (mentioned in Table 1, 2, 3, 4 and 5) using a computer based probit analysis program developed in USA (Russell *et al.*, 1977).

After the calculation of LC_{50} values of the tested insecticides due to the limitation (lack of manpower) only Diazinon 60EC was tested for hormoligosis on BPH resistance (BRRI dhan 31) and susceptible varieties (BR 3) of rice (Anonymous, 1995). For this reason BPH were reared on resistant and susceptible varieties of rice in six different iron cages. Four cages were for treatments application of hormoligant and two for control populations build-up. The BPH populations treated with hormoligant (LC_{50} and LC_{10} doses of Diazinon 60EC) were compared with that of non-treated BPH populations reared on resistant and susceptible varieties to determine the effect of hormoligant. The test populations of BPH were treated twice in each generation at 10 days interval on 3rd instar nymphs of BPH with LC_{50} and LC_{10} dosages of Diazinon 60EC. The hormoligant treated and control populations of BPH were reared on BRRI Dhan-31 and BR-3 separately in greenhouse up to 6th generation and their progenies were counted and also treated at each generation with the LC_{50} and LC_{10} dosages of Diazinon 60EC. The progenies developed in six different treatments (T_1 = LC_{50} populations of BRRI Dhan-31 (treated), T_2 = LC_{10} populations of BRRI Dhan-31 (treated), T_3 = LC_{50} populations of BR-3 (treated), T_4 = LC_{10} populations of BR-3 (treated), T_5 = Control populations of BRRI Dhan-31, T_6 = control populations of BR-3) were analyzed by one way ANOVA and means were compared by DMR test at 5% level. The LC_{50} values of BPH were tested when the treated and control populations (which reared on BRRI Dhan-31 and BR-3) reached on 5th generations.

Results and Discussion

Studies to find the effect of hormoligant (sub-lethal doses of insecticides) on brown planthopper and their LC_{50} doses were calculated during Aus and T. Aman' 1995 in laboratory and in greenhouse during 1996. Preliminary range finding tests were conducted by taking wide range of dosages. Three commonly used insecticides, Ripcord 10EC, Dimecron 100WSC, and Diazinon

Table 1: LC_{50} of Ripcord 10EC against 3rd instar nymphs of brown planthopper, *Nilaparvata lugens* (Stal)

Concentrations	Formulated doses (Lt. ha^{-1})	No. of insects tested	No. of insects dead	% Corrected mortality
0.000125	0.00062	200	198	98.91
0.0000625	0.00031	200	190	94.54
0.00003125	0.00015	200	177	87.43
0.000015625	0.000078	200	71	29.51
0.0000078125	0.000039	200	62	24.59
0.00000390625	0.000019	200	50	18.03
0.000001953125	0.0000097	200	50	18.03
Control	-	200	17	-

Table 2: LC_{50} of Ripcord 10EC against 5th instar nymphs of brown planthopper, *Nilaparvata lugens* (Stal)

Concentration	Formulated doses (Lt. /ha.)	No. of insects tested	No. of insects dead	% Corrected mortality
0.000125	0.00062	200	197	98.39
0.0000625	0.00031	200	182	90.37
0.00003125	0.00015	200	139	67.74
0.000015625	0.000078	200	146	71.12
0.0000078125	0.000039	200	46	17.65
0.00000390625	0.000019	200	34	11.23
0.000001953125	0.0000097	200	29	8.56
Control	-	200	13	-

* LC_{50} value 0.0000142 [calculated on the basis of above data by a computer program (Russel *et al.*, 1977)]

Table 3: Probit analysis for the tested dosages and their LC_{1-99} values and 95 % confidence limits for Ripcord 10EC against 3rd and 5th instar nymphs of brown planthopper

	3rd instar		5th instar	
LC[a]	Dose (%)	LCL-UCL (%)	Dose (%)	LCL-UCL (%)
1	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
10	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
20	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
30	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
40	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
50	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
60	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
70	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
80	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
90	0.0001	0.0001-0.0001	0.0001	0.0001-0.0001
99	0.0003	0.0003-0.0004	0.0003	0.0002-0.0004

LC[a]-Lethal concentration, LCL-lower 95% confidence limits and UCL- upper 95% confidence limits; Value was calculated by a computer-based program (Russell *et al.*, 1977).

Table 4: LC_{50} of Dimecron 100WSC against 5th instar nymphs of brown planthopper, *Nilaparvata lugens* (Stal)

Concentration	Formulated doses (Lt. ha^{-1})	No. of insect tested	No. of insects dead	% corrected mortality
0.003125	0.0156	100	99	98.94
0.00015625	0.00078	100	91	90.43
0.000078125	0.00039	100	81	79.79
0.0000390625	0.00019	100	68	65.96
0.00001953125	0.000097	100	61	58.51
0.000009765625	0.000048	100	35	30.85
0.0000048828125	0.000024	100	19	13.83
Control	-	100	6	-

*LC_{50} value 0.0000198722 (calculated on the basis of above data) , *Value was calculated by a computer based program (Russell *et al.*, 1977)

Table 5: LC_{50} of Diazinon 60EC against 5th instar nymphs of brown planthopper, *Nilaparvata lugens* (Stal)

Concentration	Doses (Lt. /ha.) Formulated	No. of insect tested	No. of insect dead	% Corrected mortality
0.001875	0.0093	100	100	100.0
0.0009375	0.0046	100	100	100.0
0.000046875	0.0023	100	97	96.91
0.000234375	0.0011	100	93	92.78
0.000117187	0.00058	100	36	34.02
0.000058593	0.00029	100	61	59.79
0.000029296	0.00014	100	26	23.71
0.000014648	0.000073	100	23	20.62
Control -		3	-	

*LC_{50} value 0.0000651 (calculated on the basis of above data), *Value was calculated by a computer-based program (Russell *et al.*, 1977)

Table 6: LC_{50} values of Diazinon 60EC against 5th instar nymphs of brown planthopper, *Nilaparvata lugens* (Stål)

	Mother populations			Fifth generations populations		
Treatments	LC_{50}	UCL	LCL	LC_{50}	UCL	LCL
T_1	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
T_2	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
T_3	0.0001	0.0001	0.0001	0.0001	0.0002	0.0001
T_4	0.0001	0.0001	0.0001	0.0001	0.0002	0.0001
T_5	0.0001	0.0001	0.0001	0.0001	0.0001	0.0001
T_6	0.0001	0.0001	0.0001	0.0002	0.0002	0.0002

Table 7: Development of BPH population (3rd instar) on BRRI Dhan-31 and BR-3, treated with LC_{50} and LC_{10} doses of Diazinon 60EC, Insecticide hormoligosis studies

	Populations of BPH in different generations					
Treatments	1st	2nd	3rd	4th	5th	6th
T_1	18.75c	33.3bc	92.5b	38.8c	63.ob	171.8ab
T_2	34.25bc	45.5b	90.8b	188.8a	91.3a	147.5b
T_3	91.00ab	37.3bc	75.0b	200.0a	65.5b	224.5a
T_4	90.00ab	88.5a	78.3b	98.8b	98.8a	225.0a
T_5	107.5ab	16.5c	170.0a	30.0c	30.0c	32.5c
T_6	126.25a	28.0bc	50.5b	65.0b	65.0b	235.3a

T_1 = LC_{50} populations of BRRI Dhan-31 (treated), T_2 = LC_{10} populations of BRRI Dhan-31 (treated), T_3 = LC_{50} populations of BR-3 (treated), T_4 = LC_{10} populations of BR-3 (treated), T_5 = Control populations of BRRI Dhan-31, T_6 = Control populations of BR-3. Means in each column followed by the same letter are not significantly different at 5% level by DMRT.

60EC were tested for this purpose. The tested doses and their respective responses (% corrected mortality) against different concentrations of different insecticides were calculated (Table 1, 2, 3, 4 and 5). Probit analysis showed that the LC_{50} value of Ripcord 10EC both for 3rd and 5th instar nymphs of BPH was 0.0000142% (Tables 1, 2 and 3). The LC_{50} value of Dimecron 100WSC and Diazinon 60EC for 5th instar nymphs of BPH were 0.00001987% (approx. 0.00009935 Lt ha^{-1}) and 0.000651% (approx. 0.00005425 Lt ha^{-1}) (Tables 4 and 5).

To find the effect of hormoligant (sub lethal dosages of insecticide i.e LC_{10}) on brown planthopper in resistant (BRRI Dhan-31) and susceptible (BR 3) varieties were conducted in greenhouse. But the LC_{50} values of mother and 5th generation's populations had no change (Table 7). Yoshioka and Yamaskai (1981) tested the effect of selection on the development of resistance of *Nephotettix cincticeps* in the laboratory with a combination of iprobenfos and malathion, LD_{50} values of the mixture only increased 1.5 times by the F19 generation; by F26, the LD_{50} had increased seven fold. Actually to find the effect of hormoligant, these studies needed more years of trials.

But the effect of hormoligant (i.e. the effect of LC_{50} and LC_{10} dosages of Diazinon 60EC on the test populations) was observed in 1st, 2nd, 5th, and 6th generations of the treated populations; i.e. the populations in LC_{10} in BPH resistant and susceptible varieties increased at the above generations compared to LC_{50} populations of BPH (Table 7). Similarly, in a preliminary study at BRRI, 3 commercial insecticides i.e. Diazinon 60EC (diazinon), Azodrin 40 WSC (monocrotophos), Lebaycid 50EC (malathion) were found to cause higher fecundity when applied at 0.04% concentration using 1000 litre spray volume per hectare. This means that these insecticides may cause BPH resurgence (Alam and Karim, 1986). Chelliah (1980) found that the BPH populations were highest on plots sprayed 3 times with deltamethrin. Chelliah and Heinrichs (1984) also reported that the feeding rate of *N. lugens* have increased at sub lethal dosages of resurgence inducing insecticides applications. Similarly, Kerns and Stewart (2000) reported that the sub lethal effects of dosages of bifenthrin on aphids by some increases in the net reproductive rate. Increased nymph production has been found in the progeny of *Myzus persicae* exposed to plants sprayed with the organophosphorus insecticides (azinophosmethyl) possibly as a result of improved nutrition or an effect on the reproductive hormones in the mother

(Coombes, 1983). Buenaflor *et al.* (1981) found that when certain insecticides (deltamethrin) are applied to rice varieties susceptible (TN 1) to the brown planthopper (*N. lugens*), a resurgence of the planthopper

populations commonly occurs. But in this experiments the populations of brown planthopper also increased in resistant variety (BR 31) (Table 7); so further studies were needed to confirm these findings.

To save money, farmers are using low insecticides doses. This practice, combined with the short residual toxicity of many commercial insecticides, will often cause the BPH to be exposed to sublethal insecticide doses, low doses of resurgence-inducing insecticides increased the reproductive rate of the BPH and reduced the nymphal duration, eventually leading to resurgence (Chelliah and Heinrichs, 1984; Anonymous, 1984).

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