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Antibiosis effects of selected rice genotypes on the brown planthopper, *Nilaparvata lugens* (Stal.)

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ABSTRACT

Aim: The aim of the study was to evaluate the reaction of several rice genotypes and for their antibiosis effects on BPH, focusing mainly on key biological parameters such as nymphal survival, developmental duration, and feeding activity.

Materials and Methods: A total of 14 different rice genotypes collected from different countries were selected and evaluated along with resistant check, Ptb 33 and susceptible check, TN1 for their resistance or susceptibility to BPH populations of Godavari Delta and for antibiosis studies under polyhouse conditions.

Results: Antibiosis parameters of resistance *viz*., nymphal survival, nymphal duration, growth index and feeding rate were also evaluated in all rice genotypes. Among those, RP 2068-18-3-5 exhibited highly resistant reaction (1.0) and five other cultures *viz*., resistant check, Ptb 33, IR 36, Chinsaba, Pokkali and IR 7033-121-15 were showed resistant reaction (1.0 to 3.0). Low survival rate was recorded on RP 2068-18-3-5 (13.30%) and Chinsaba (23.33%). Whereas, prolonged development period of nymphs was recorded in Ptb33 (20.67 days) and was followed by IR 7033-121-15 (14.33 days), IR36 and RP 2068-18-3-5 (15.0 days each). Low growth index value was observed on RP 2068-18-3-5 (0.88) and was followed by Chinsaba (1.94) and resistant check, Ptb 33 (1.94). The rice genotypes i.e. Ptb 33 (67.33 mm^2), RathuHeenati (100.67 mm^2), IR36 (151.33 mm^2) and MUTNS-1 (246.67 mm^2) recorded lowest honeydew excreted area. The rice genotypes with resistant genes Bph3 (Ptb33) and Bph 2 (IR36) showed highly resistance reaction and also confirmed its high level of antibiosis to BPH population in Godavari delta.

Conclusion: It was concluded that the rice genotypes with different BPH-resistant genes exhibited different levels of antibiosis.

Keywords: Rice genotypes, Brown planthopper, Reaction, Mechanisms of resistance, Antibiosis.

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Introduction

Rice (*Oryza sativa* L.) is one of the most important staple food crops, feeding more than half of the global population. It is grown in more than 100 countries with 90% of total global production from Asia (Fukagawa and Ziska, 2019). Out of 782 million tons of global rice production from 167.1 million hectares, India produced 116.42 million tons in 44.5 million hectares (FAO, 2020). However, its production is severely threatened by a range of insect pests, among which brown planthopper (BPH), *Nilaparvata lugens* Stål is considered one of the most destructive especially in Asia-Pacific region (Iamba and Danar, 2021). BPH not only causes direct damage by feeding on phloem sap, resulting in hopper burn, but also acts as a vector for viral diseases such as rice ragged stunt and grassy stunt viruses.

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Presently, management of BPH mainly relies on chemical pesticides, though this approach is costly, resulting in environment contamination, harmful effects on non-target organisms, and also increased resistance and resurgence in planthoppers (Wu *et al*., 2020). An alternative most economical and environmentally safe strategy for effective management of this insect pest is cultivation of resistant varieties (Sarao *et al*., 2016). Therefore, it is very important to identify resistant sources in rice genotypes with different mechanisms of resistance for the development of resistant varieties against BPH.

Host-plant resistance employs various mechanisms i.e. non-preference, antibiosis and tolerance (Painter, 1951). Among those, antibiosis plays a crucial role in suppressing pest population growth by adversely affecting the biology and development of the insect. Antibiosis resistance may manifest through delayed development, reduced survival, lower

fecundity, or impaired feeding of the pest (Wang *et al*., 2018). Understanding and identifying rice genotypes that exhibit strong antibiosis effects against BPH is therefore critical for developing resistant rice varieties for sustainable and environment-friendly management of BPH. This study aims to evaluate the reaction of several rice genotypes and for their antibiosis effects on BPH, focusing mainly on key biological parameters such as nymphal survival, developmental duration and feeding activity.

Materials and Methods

The present investigations were carried out under polyhouse conditions at Andhra Pradesh Rice Research Institute and Regional Agricultural Research Institute, Maruteru, West Godavari district, Andhra Pradesh during the year 2014-15.

Mass culturing of BPH

Brown planthopper was mass reared at 30°±5°C on potted plants of TN1 (Taichung native1) variety (Heinrichs *et al*., 1985). The newly emerged first to fifth instar nymphs of BPH and female adults were utilized for screening and antibiosis studies.

Plant material

A total of 14 different rice genotypes collected from different countries were selected and evaluated along with resistant check, Ptb 33 and susceptible check, TN1 for their resistance or susceptibility to BPH populations of Godavari Delta and for antibiosis studies under polyhouse conditions (Table 2).

Standard Seed box screening technique

The pre germinated seeds of test cultures were sown 3 cm apart in a seed box. Each test culture was sown in three replications in a row across the width of the seed box with at least 20 plants per row. At seven days after sowing, the seedlings were infested with second and third instar nymphs of BPH at rate of eight to ten nymphs per seedling. After infestation each seed box was covered with a wire mesh cage to prevent any escape and to prevent entry of natural enemies. Test cultures were observed daily for damage by BPH. Damage rating of test cultures was observed when 90 per cent of seedlings in TN1 were killed. The test cultures were graded using standard evaluation system (SES) of rice on 0-9 scale as given (Table 1) (IRRI, 1996).

Evaluation of antibiosis parameters in rice genotypes

The experiments on antibiosis *viz*., nymphal survival, nymphal development period, growth index and feeding rate were conducted. To conduct these antibiosis studies the seeds of each test culture along with resistant and susceptible checks were sown in an earthen plastic pot individually and at fifteen days after sowing, 2-3 seedlings of each test culture were transplanted in 10-cm diameter plastic pots and were then covered with cylindrical Mylar film cages to protect from other pests and predators.

Feeding rate

The antibiosis resistant to BPH in rice was assessed by estimating the amount of honeydew excreted by the female adult hoppers. Whatman No.1 filter paper was dipped in a 0.02% bromocresol green solution in ethanol was placed around the base of the feeding chamber and infested with five freshly emerged female BPH hoppers, pre-starved for four hours. The BPH adults were allowed to feed for 24 hours at the base of the stem. Honeydew excreted by BPH was absorbed by the filter paper and appeared as a blue spot. Area of the spot was proportional to the honeydew production and therefore to the amount of BPH feeding (Heinrichs *et al*., 1985).

Nymphal survival

Rice genotypes tested along with resistant and susceptible checks grown separately in plastic pots (10 cm diameter) and were infested with ten first instar BPH nymphs on 30-days old Mylar sheet caged plants. The top open end of the tube was closed with muslin cloth to prevent migration. Each culture was replicated five times.

Table 1. Standard Evaluation System (SES) describing the score of plant based on its reaction to BPH incidence

Grade	Criteria	Reaction	Consideration of damage score
0	No damage	Immune (I)	0
1	Very slight yellowing	Highly resistant (HR)	0.1 to 1.0
3	First and 2nd leaves of most of plants partially yellowing	Resistant (R)	1.1 to 3.0
5	Pronounced yellowing and stunting or about 10 to 25 % of the plants wilting	Moderately resistant (MR)	3.1 to 5.0
7	More than half of the plants wilting or dead and remaining plants severely stunted or drying	Moderately susceptible (MS)	5.1 to 7.0
9	All plants dead	Highly susceptible (S)	7.1 to 9.0

At 20 days after infestation, counted the number of live hoppers on rice genotype and calculated the survival percentage of nymphs (Heinrichs *et al*., 1985) to compare their antibiosis effect on nymphs.

Nymphal development period

Nymphal development period on rice genotypes along with resistant and susceptible checks was studied by releasing five first instar BPH nymphs on 30-days old Mylar film caged plants. From 9th day onwards, nymphs on each culture were observed daily for ecdysis and recorded the number of days taken for the nymphs to reach adult stage on each rice genotype (Pongprasert and Weeraput, 1979).

Growth Index (GI)

Growth index of BPH on each rice genotype and the resistant and susceptible checks was computed by using the data obtained from the experiments on nymphal survival and development period (Panda and Heinrichs, 1983) as follows:

$$\text{Growth Index} = \frac{\text{\% nymphs survived on test culture}}{\text{Development period of nymphs on test culture}}$$

The data obtained from the experiments were subjected to Completely Randomized Design (CRD) using analysis of variance (ANOVA) after transforming the data into suitable transformations and the mean values were compared (Gomez and Gomez, 1984).

Results and Discussion

Reaction of rice genotypes to BPH population of Godavari delta: A total of fourteen rice cultures screened against BPH in comparison with susceptible check (TN1) and resistant check (Ptb 33) in polyhouse through standard seed box screening test and the results were presented (Table 2). The results indicated that there were significant differences among rice genotypes in damage score against brown planthopper. Out of those,one culture was highly resistant *viz*., RP 2068-18-3-5 (Score 1.0); five cultures *viz*., resistant check, Ptb 33 (Score 1.4), IR 36 (Score 1.9), Chinsaba (2.3), Pokkali (2.8) and IR 7033-121-15 (3.0) are resistant and six were moderately resistant (Babawee, Swarnalatha, Rathu Heenati, T12, IR 65482-7-216-1-2-B and OM 4498 with a score ranging from 3.1 to 5.0. The genotypes Milyang 63 (Score 7.0), IR 64 (score 7.3), MUTNS-1 (Score 8.7) and susceptible check TN1 (Score 9.0) showed susceptible reaction.

Feeding rate

The quantity of excretion of honeydew by brownplanthopper, in general, is directly related to intake of plant sap. Therefore, the amount of honeydew excreted by the insect in unit time when fed on different rice genotypes is considered as an index for its feeding preference. The honeydew excreted area of BPH adults in mm^2 on selected rice genotypes in comparison to the susceptible and resistant check were presented (Table 3). The amount of honeydew excreted in the present study ranged from 67.33 mm^2 to 983.33 mm^2. The results indicated that there was a significant difference among the rice genotypes in amount of honeydew excretion. Among the resistant rice genotypes, the resistant check, Ptb 33 recorded lowest honeydew excreted area of 67.33 mm^2 and was on par with Rathu Heenati (100.67 mm^2) and were followed by IR36 (151.33 mm^2), MUTNS-1 (246.67 mm^2). The rice genotypes which recorded moderate honeydew excreted area were RP 2068-18-3-5 (307.00 mm^2), T12 (344.00 mm^2), Swarnalatha (359.00 mm^2). The honeydew excreted area of rice genotypes *viz*., OM44982 (517.33 mm^2), Babawee (491.67 mm^2), IR65482-7-216-1-2-B (537.67 mm^2), IR 7033-121-15 (561.33 mm^2), Chinsaba (558.00 mm^2), Pokkali (735 mm^2) and IR 64 (776.67 mm^2) were on par with susceptible check, TN1 (440.00 mm^2). The rice genotype, Milyang 63 recorded significantly highest honeydew area (983.33 mm^2) among all the rice genotypes tested.

Nymphal survival

The data on the per cent survival of BPH nymphs on the fourteen selected rice cultures along with the resistant and susceptible checks indicated wide differences in the survival pattern of BPH nymphs (Table 3). The nymphal survival after 20 days of their release on the tested rice cultures was ranged from 13.33 to 100.00 per cent. Among the rice cultures, the lowest per cent of nymphal survival was observed on highly resistant culture, RP 2068-18-3-5 (13.30%) and was on par with Chinsaba (23.33%). These were followed by IR 7033-121-15 (36.67%), RathuHeenati (40.0%), resistant check, Ptb 33 (40.0%), T12 (43.33). The rice cultures with moderate % of nymphal survival were Babawee (50.0%), Milyang 63 (53.33%), IR65482-7-216-1-2-B (56.67%), IR36 (56.67%), Pokkali (60.0%).

Table 2. Reaction of rice genotypes with different resistant genes against BPH in seed box screening test during 2014

S. No.	Designation	Origin	Resistant gene	Damage Score*	Reaction
1	RathuHeenati	Sri Lanka	Bph3, Bph14, Bph17	3.2	MR
2	Babawee	Sri Lanka	Bph4	5.0	MR
3	Milyang 63	Republic of Korea	Bph6	7.0	MS
4	Swarnalatha	India	Bph6	3.9	MR
5	T12	Thailand	bph7	4.0	MR
6	Chinsaba	Myanmar	bph8	2.3	R
7	Pokkali	Sri Lanka	Bph9	2.8	R
8	IR 64	Philippines	Bph1	7.3	S
9	IR65482-7-216-1-2-B	Australia	Bph18	4.8	MR
10	IR 36	Philippines	Bph2	1.9	R
11	MUTNS-1	India	Bph35	8.7	HS
12	OM4498	Vietnam	Bph45	4.0	MR
13	RP 2068-18-3-5	India	Bph33	1.0	HR
14	IR 7033-121-15	Philippines	Bph20/21	3.0	R
15	Ptb 33	India	Bph2+Bph3	1.4	R
16	TN1	Taiwan	---	9.0	HS
	F test	---	---	Sig	---
	CD (0.05)	---	---	0.23	---
	CV (%)	---	---	3.96	---

*Mean of 10 individual plants and three replications

Table 3. Antibiosis effects of rice genotypes on honeydew excretion, % nymphal survival, nymphal development period and growth index of BPH during2014-15

S. No.	Designation	Reaction	Honeydew excretion (mm²) *	% nymphal survival**	Nymphal development period (days)	Growth index
1	RathuHeenati	MR	100.67 (9.89)	40.00 (38.85)	13.33	3.04
2	Babawee	MR	491.67 (22.05)	50.00 (45.00)	12.67	3.97
3	Milyang 63	MS	983.33 (30.85)	53.33 (46.92)	12.00	4.44
4	Swarnalatha	MR	359.00 (18.44)	56.67 (48.93)	10.67	5.27
5	T12	MR	344.00 (18.51)	43.33 (41.07)	12.00	3.61
6	Chinsaba	R	558.00 (23.59)	23.33 (28.08)	12.00	1.94
7	Pokkali	R	735.00 (27.06)	60.00 (50.85)	12.00	5.00
8	IR 64	S	776.67 (27.84)	70.00 (57.00)	13.00	5.43
9	IR65482-7-216-1-2-B	MR	537.67 (23.14)	56.67 (48.85)	12.67	4.48
10	IR 36	HR	151.33 (12.07)	56.67 (48.93)	15.00	3.82
11	MUTNS-1	HS	246.67 (15.39)	93.33 (81.14)	11.67	7.98
12	OM4498	MR	517.33 (21.61)	63.33 (52.86)	12.33	5.17
13	RP 2068-18-3-5	HR	307.00 (17.29)	13.33 (21.14)	15.00	0.88
14	IR 7033-121-15	R	561.33 (23.48)	36.67 (37.14)	14.33	2.54
15	Ptb 33	HR	67.33 (9.89)	40.00 (39.15)	20.67	1.94
16	TN1	HS	444.0 (22.05)	100.0 (90.0)	11.33	8.83
	CD (0.05)		3.65	7.21	0.67	0.85
	CV (%)		14.43	11.75	4.06	15.85

*Figures in parenthesis are square root transformed values
**=Figures in parenthesis are Arcsine transformed values

Significantly highest per cent of nymphal survival was observed in OM4498 (63.33%), IR64 (70.0%), MUTNS-1 (93.33%) and susceptible check, TN1 (100.0%)

Nymphal development period

Nymphal development period in days was significantly prolonged on the resistant and moderately resistant genotypes (Table 3). Nymphal period ranged between 10.67 to 20.67 days. Significantly prolonged development period was observed in resistant check, Ptb33 (20.67 days) and it was followed by IR 7033-121-15 (14.33 days), IR36 and RP 2068-18-3-5 (15.0 days each). These were followed by RathuHeenati (13.33 days) and was on par with IR64 (13.0 days), Babawee, IR65482-7-216-1-2-B (12.67 days each), and OM4498 (12.33 days). The other genotypes *viz*., Chinsaba, Pokkali, T12 and Milyang 63 and MUTNS-1 recorded a greater number of days to complete nymphal duration than TN1 (11.33 days). Whereas, Swarnalatha took lesser number of days (10.67) than TN1.

Growth index

The growth index values for the tested rice genotypes were presented in table 3. The values of growth index ranged from 0.88 to 8.84. All the rice genotypes registered significantlylow growth index than the susceptible check TN1 (8.84). Among the rice genotypes, significantly lowest growth index was observed in RP 2068-18-3-5(0.88) and was followed by Chinsaba (1.94) and resistant check, Ptb 33 (1.94). These were followed by IR 7033-121-15 (2.54) and was on par with T12 (3.61), IR36 (3.82). The other rice genotypes *viz*., Babawee (3.97), IR65482-7-216-1-2-B (4.48), Pokkali (5.0), OM44982 (5.17), Swarnalatha (5.27) and IR64 (5.43) registered moderate growth index. The highest growth index was observed in MUTNS-1 (7.78) but was significantly lower than TN1 (8.84).

In the present investigation, the rice varieties *viz*., Pokkali was resistance, Swarnalatha and T12 were moderately resistance. These findings were partially in agreement with the findings of Qiu *et al*. (2011), who reported Pokkali and Swarnalatha as highly resistant; T 12 as moderately resistant. This might be due to difference in BPH biotypes and their virulence levels to rice genotypes carrying different resistance genes. The rice genotype with resistant gene, Bph 33 (RP 2068-18-3-5) exhibited highly resistant reaction. The other rice genotypes with Bph3 (Ptb33), Bph 2 (IR36) and Bph20/21 (IR 7033-121-15), bph8 (Chinsaba), Bph9 (Pokkali) showed resistant reaction and the rice genotypes with Bph4 (Babawee), Bph6 (Swarnalatha), bph7 (T12), Bph18 (IR65482-7-216-1-2-B), Bph45 (OM4498) and Bph17/18 (RathuHeenati) genes showed moderately resistant reaction to Godavari population of BPH. Whereas, IR64 (Bph1), Milyang 63 (Bph6) and MUTNS-1 (Bph35) exhibited susceptibility reaction (Table 2).

In the present study evaluated the antibiosis effects of selected rice genotypes on BPH through per cent nymphal survival, development period, growth index, and feeding rate. The results demonstrated significant differences among the tested genotypes, indicating varying levels of antibiosis mediated resistance. The variation in per cent nymphal survival among genotypes suggests a strong influence of host plant quality on the survivability of BPH nymphs. Genotypes exhibiting lower nymphal survival rates, such as RP 2068-18-3-5, Chinsaba, IR7033-121-15, Rathu Heenati, Ptb 33 and T12 could be considered to possess high antibiosis potential. Reduced survival may be attributed to the presence of feeding deterrents, such as soluble salicylic acid, malic acid, itaconic acid and benzoic acid or inadequate nutrients or absence of essential nutrients in the resistant genotypes as reported by Soundararajan *et al*. (2003).

The Prolonged development periods observed on resistant genotypes i.e. Ptb33, IR36, RP 2068-18-3-5 and IR 7033-121-15 in the present study indicate the presence of inhibitory factors that delay nymphal maturation. Extended development period not only reduce the reproductive potential of BPH populations but also expose them to natural enemies and environmental hazards for a longer duration under field conditions. This effect further supports the role of antibiosis in suppressing pest buildup. The growth index, which reflects both survival and development, was significantly lower in resistant genotypes like RP 2068-18-3-5, Chinsaba and resistant check, Ptb 33, further confirming antibiosis as a mechanism of resistance. Genotypes with low growth indices likely interfere with hormonal regulation, nutrient assimilation, or cellular metabolism of the insect, thereby limiting successful development. Significantly reduced feeding rates on resistant genotypes like Ptb 33, RathuHeenati, IR36 and MUTNS-1 suggest that these rice varieties are less palatable to BPH may be due to presence of some biochemical deterrents that hinder the insect's ability to effectively utilize phloem sap. This not only limits direct damage to the host plant but also weakens the insect due to nutritional deficiency. Similar findings of low per cent nymphal survival**,** lower growth index and honeydew excretion in resistant genotypes like RP 2068-18-3-5 (Anita *et al*., 2014), MTU IJ 206-7 4-1 (Bhanu *et al*., 2014b), CR3006-8-2, RP4918-221, KAUM182-1 (Singh *et al*., 2017) and RathuHeenati (Li *et al*., 2022) than susceptible check, TN1were also reported earlier.

The expression of the mechanisms of resistance is also governed by the associated resistance genes to BPH. In the present study, Ptb 33 and IR 36 with Bph2 resistant gene recorded prolonged development period clearly indicated that this gene may confers antibiosis mechanism of resistance on nymphal development period. Similarly, Ptb 33 and Rathu Heenati with Bph3 gene recorded lesser honeydew excreted area indicated that this gene

Table 4: Rice genotypes with different antibiosis indices along with their resistant genes against brown planthopper

S. No.	Antibiosis indices	Rice genotypes (Resistant genes)
1	Honeydew excreted area (mm^2)	Ptb 33 (Bph3), Rathu Heenati (Bph3), IR36 (Bph2), MUTNS-1 (Bph35), RP 2068-18-3-5 (Bph33), T12 (bph7) and Swarnalatha (Bph6).
2	Nymphal survival (%)	RP 2068-18-3-5 (Bph33), Chinsaba (bph8), IR 7033-121-15(Bph20/21), Rathu Heenati (Bph3), Ptb 33 (Bph2) and T12 (bph7).
3	Nymphal duration (days)	Ptb33(Bph3), IR 7033-121-15 (Bph20/21), IR36 (Bph2) and RP 2068-18-3-5 (Bph33).
4	Growth index	RP 2068-18-3-5(Bph33), Ptb 33 (Bph2 and 3), IR 7033-121-15(Bph20/21), T12 (bph7) and IR36 (Bph2).

is responsible for antibiosis on feeding. RP 2068-18-3-5 (Bph33), Chinsaba (bph8) and IR 7033-121-15 (Bph20/21) were also showed high level of antibiosis on nymphal survival, development period. The other rice genotypes with different resistant genes *viz*., Swarnalatha (Bph6), T12 (bph7), Pokkali (Bph9), IR65482-7-216-1-2-B (Bph18) and OM4498 (Bph45) exhibited moderate level of antibiosis on nymphal survival, development period indicated that these resistant genes are responsible for any one of the different mechanisms of resistance against BPH (Table 4). Bhanu *et al*., 2014a and Tammarai and Soundararajan, 2017 were also reported earlier similar results.

Conclusions

It was concluded that the rice genotypes with different BPH-resistant genes exhibited different levels of antibiosis. Overall, the results reinforce the importance of antibiosis as a mechanism of resistance in rice against BPH. The findings will contribute to the identification of potential donor lines with effective antibiosis resistance, which can be utilized in rice breeding programs for durable BPH resistance. Genotypes like Ptb33, RP 2068-18-3-5, IR36, RathuHeenati and IR 7033-121-15 demonstrated strong antibiosis traits in the present study. Furthermore, the varieties developed through the use of these genotypes' may also be integrated with other control strategies such as biological control and minimal insecticide use could lead to more sustainable and eco-friendlier BPH management. Future research should focus on identifying the specific biochemical and morphological traits responsible for antibiosis and validating them under field conditions.

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