

Journal of Advances in Biology & Biotechnology

Volume 27, Issue 8, Page 442-452, 2024; Article no.JABB.120142 ISSN: 2394-1081

# Biochemical Defense Responses in Advanced Backcross-derived Rice Genotypes to *Nilaparvata lugens* (Stal) Infestation

R. Prathima Kumari <sup>a\*</sup>, Ch. Anusha <sup>b++</sup>, M. A. Prajwal Gowda <sup>a</sup>, D. Akhilandeshwari <sup>a</sup>, R. Shravan Kumar <sup>c#</sup>, K. Rajendra Prasad <sup>d++</sup> and Y. Hari <sup>e#</sup>

<sup>a</sup> Department of Entomology, College of Agriculture, PJTSAU, Rajendranagar, Hyderabad, India.
 <sup>b</sup> Department of Entomology, Agricultural College, Warangal, PJTSAU, India.
 <sup>c</sup> Rice Section, Regional Agricultural Research Station, Warangal, PJTSAU, India.
 <sup>d</sup> Department of Genetics and Plant Breeding, Agricultural College, Warangal, PJTSAU, India.
 <sup>e</sup> Biotechnology Department, Regional Agricultural Research Station, Warangal, PJTSAU, India.

## Authors' contributions

This work was carried out in collaboration among all authors. 'All authors read and approved the final manuscript.

#### Article Information

DOI: https://doi.org/10.9734/jabb/2024/v27i81156

#### **Open Peer Review History:**

This journal follows the Advanced Open Peer Review policy. Identity of the Reviewers, Editor(s) and additional Reviewers, peer review comments, different versions of the manuscript, comments of the editors, etc are available here: https://www.sdiarticle5.com/review-history/120142

> Received: 17/05/2024 Accepted: 19/07/2024 Published: 23/07/2024

Original Research Article

++ Assistant Professor;

**Cite as:** Kumari, R. Prathima, Ch. Anusha, M. A. Prajwal Gowda, D. Akhilandeshwari, R. Shravan Kumar, K. Rajendra Prasad, and Y. Hari. 2024. "Biochemical Defense Responses in Advanced Backcross-Derived Rice Genotypes to Nilaparvata Lugens (Stal) Infestation". Journal of Advances in Biology & Biotechnology 27 (8):442-52. https://doi.org/10.9734/jabb/2024/v27i81156.

<sup>#</sup> Scientist;

<sup>\*</sup>Corresponding author: E-mail: prazzzugowdru@gmail.com;

## ABSTRACT

BPH, Nilaparvata lugens (Delphacidae: Homoptera) infestation on most of the resistant and moderately resistant backcross derived rice genotypes and resistant checks (PTB 33 and BM 71) resulted in an increase in the phenolic content in the infested plants leaf sheath. The percentage increase in phenols in resistant and moderately resistant genotypes ranged from 14.8 % to 67.7 %. Upon infestation of BPH there was a decrease in total reducing sugars in all the resistant and moderately resistant genotypes. The highest quantity of reducing sugars was present in healthy susceptible check TN1 compared to all the resistant and moderately resistant backcross derived rice genotypes. The ascorbic acid content in the resistant and moderately resistant genotypes decreased after BPH infestation and percentage decrease ranged from 16% to 36%. The total N content in the infested resistant and moderately resistant genotypes decreased over healthy genotypes. Decrease in the N content was highest in the susceptible check TN1 (35.2%). In the resistant checks slight decrease in the N content was observed. Potassium and Phosphorous (%) increased in the resistant genotypes and resistant checks, but not in susceptible check TN1. The plants responded defensively upon infestation, resulting in production of higher amount of phenolics, potassium, phosphorous and reduced level of nitrogen, reducing sugars and ascorbic acid.

Keywords: BPH; phenol; ascorbic acid; nitrogen.

#### **1. INTRODUCTION**

As a major staple crop, Oryza sativa (rice) is vital for maintaining global food security [1]. It is grown in 114 countries and acts as the main source of income for over 100 million households in Asia and Africa. Rice, a member of the Gramineae family, possesses a genome size of 430 MB. Globally, it is cultivated across 162.06 million hectares, yielding 500 million metric tons annually, averaging 5.0 tons per hectare [2]. India is the second-largest rice-producing country, next only to China. Rice cultivation in India covers 45.5 million hectares, producing 125 million metric tons annually at an average yield of 4.1 metric tons per hectare [3]. Nearly, 52% of global rice production is lost due to biotic stresses, with insect pest attacks accounting for 25% of these losses [4]. Atwal and Dhaliwal [5] reported that of the over 100 insect species that affect rice, 20 are categorized as major pests. Of all, Nilaparvata lugens, is considered a notorious pest in Asia for its damaging impact on rice crops due to its feeding habits on phloem sap [6]. Nymphs and adults feeding on sap from leaves and leaf sheaths lead to symptoms such as vellowing leaves, reduced tillering, shorter plant height, and increased grain unfilledness. An extensive infestation of BPH leads to a condition termed 'hopper burn' [7]. Host plant resistance is a fundamental approach that is an economically viable and advisable tactic in BPH management. Biochemical constituents of the rice plant contributing to resistance has been studied from time immemorial. Here, there are a total of 15

includina advanced aenotypes. backcrossderived genotypes and resistant and susceptible checks. Siddhi backcross-derived genotypes (12) of F6 generation were used. The genotypes were categorized into resistant, moderately resistant, and susceptible based on our research conducted under both field and glasshouse [8]. However, the samples for conditions biochemical parameters were taken from glasshouse grown plants. Least square difference (LSD) test was used to compare the differences in mean. Hence, the current study aimed to identify how certain biochemical parameters impart resistance against BPH.

## 2. MATERIALS AND METHODS

#### 2.1 Experimental Area

In *kharif* 2021, this research was conducted at the RARS in Warangal to understand the biochemical defense responses that might be present in the previously screened advanced backcross-derived rice genotypes [8].

#### **2.2 Estimation of Phenols**

The phenol content present in 1g of leaf sheath was estimated by Folin-ciocalteau method (Singleton et al. 1999).

**Reagents:** Gallic acid: 100 mg gallic acid was dissolved in 100 ml distilled water for 1000 ppm. Working standard: 1 ml of stock added in 20 ml water for 50 ppm concentration.

#### Procedure:

- A. To 1gm of sample, 10 ml of methanol was added in centrifuge tubes and kept it for maceration for 24hrs.
- B. Samples were centrifuged at 6000 rpm for 15 min, taken into 10 ml volumetric flask through filtering and made upto mark with methanol. This extract was used for phenols estimation.
- C. After making up the volume to 3.0 ml with distilled water, 1 ml of Folin-Ciocalteau reagent, 2.0 ml of 20% Na2CO3 were added and tubes were placed in a boiling water bath for 1 min. After allowing the content of tubes to cool, the blue solution was diluted to 25 ml with distilled water, the colour developed was measured at 650 nm against a reagent blank in a spectrophotometer.
- D. The concentrations of total phenol present in the unknown sample were extrapolated from the standard calibration curve. The amount of total phenol present in the leaf sheath was expressed as mg g<sup>-1</sup>.

**Estimation of reducing sugars:** Total Reducing sugars present in rice leaf sheath was estimated by Nelson-Somyogi method [9].

## **Reagents:**

i Alkaline copper tartarate reagent

**Reagent A:** 2.5g of anhydrous sodium carbonate, 2 g of sodium bicarbonate, 2.5 g of sodium.

Potassium tartarate, 20 g of sodium sulphate dissolved in 80 ml distilled water and made upto 100 ml.

**Reagent B:** 15 g of copper sulphate was dissolved in 20 ml distilled water, one drop of concentrated sulphuric acid is added and made upto 100 ml.

**Reagent C:** 4 ml of B and 96 ml of A solution mixed before use.

ii. Arsenomolybdate reagent

2.5 g of ammonium molybdate was dissolved in 45 ml of distilled water. 2.5 ml of sulphuric acid was added and mixed well. Then 0.3 g of disodium hydrogen arsenate was dissolved in 2.5 ml of water.

- iii. 80% ethanol 20 ml of water was added to 80 ml of ethanol.
- iv. Glucose standard 100 mg of glucose was dissolved in 100 ml of distilled water for obtaining 1000 ppm.

#### Procedure:

- A. 100 mg of sample was taken into centrifuge tubes and sugars were extracted by adding hot 80% ethanol (twice) 5ml each time.
- B. Supernatant was collected into another centrifuge tubes and evaporated it by keeping it on a water bath at 80°C.
- C. Sugars were dissolved by adding 10 ml of water.
- D. A set of the test tubes containing 0.2, 0.4, 0.6, 0.8 and 1ml. of standard solution were prepared.
- E. 1 ml of aliquot was added into test tubes using pipette. The volume of aliquots in all the test tubes were made up to 2.0 ml with distilled water.
- F. Later, 1 ml of alkaline copper tartarate was added to test tubes and were placed in boiling water for 10 min.
- G. The test tubes were cooled and 1 ml of arsenomolybdic acid reagent was added.
- H. 6 ml of distilled water was added to test tubes and the absorbance (intensity of the colour) was read in a spectrophotometer at 620 nm. The standard graph was drawn from the absorbance values and the amount of sugars was calculated from the standard graph. The amount of sugar present in the sample was expressed as mg g<sup>-1</sup> of plant tissue.

**Estimation of ascorbic acid:** Total amount of ascorbic acid present in rice leaf sheath was estimated by Direct Calorimetric determination method described by Sadasivan and Manickam [10].

#### Reagents:

a) 2% metaphosphoric acid: 32 g of metaphosphoric acid added to1600 ml of water and made upto 2000 ml. b) Dye solution: 50 mg of 2,6-dichlorophenol indophenol and 42 mg of sodium bicarbonate dissolved in 50 ml hot distilled water. Filtered and 25 ml was diluted to 500 ml with distilled water. c) Standard stock solution: 100 mg ascorbic acid was dissolved in 100 ml of 4% metaphosphoric acid. d) Working

standard solution: 5ml of stock was taken into 50 ml volumetric flask and made upto mark.

#### Procedure:

- a. 5 g of sample was taken in centrifuge tubes.
- b. 25 ml of 2% metaphosphoric acid was added and homogenized for 10 min.
- c. 25 ml of 2% metaphosphoric acid was then made upto 50 ml with metaphosphric acid and centrifuged for 15 min at 4000 rpm.
- d. Readings were taken at 518 nm using spectrophotometer.

**Estimation of nitrogen:** Nitrogen was estimated on whole plant basis using the standard microkjeldhal method Zuazaga and Ma, [11] and expressed as percentage.

**Procedure:** Digestion: 0.2 g of sample was taken and 3 g of catalyst mixture (40 g of potassium sulphate and 8 g of copper sulphate) was added to it. Sample was digested by adding 10 ml of concentrated sulphuric acid at 42°C for 2 hrs 30min. Distillation was done with 4% NaOH, 0.4% Boric acid and titrated with 0.1N sulphuric acid for nitrogen percentage.

**Estimation of Phosphorous:** Phosphorus content was determined by following Piper, [12] method. Bortom's reagent :11.5 g of ammonium molybdate was dissolved in 200 ml of water. 0.62 g of ammonium vanadate was dissolved in 300 ml of water. Two solutions were added in 500 ml volumetric flask. 125 ml of nitric acid was added to it and made upto mark with distilled water.

**Procedure:** 2.5 ml of extract was taken into 25 ml into volumetric flask. 5 ml of Bartom's reagent was added and made upto mark with distilled water. Phosphorous was determined with the help of spectrophotometer at 420 nm after 30 min.

#### Estimation of Potassium:

Potassium content was determined by following Piper, [12] method. Sample preparation: Plant material was digested by wet digestion method [12] using diacid mixture (nitric and perchloric acids in 9:4 ratio). 1 g of plant sample was taken in conical flask and 12-15 ml of diacid mixture was added. Later it was kept for digestion at 200°C until clear colour appears on hot plate. After cooling 20 ml of water was added and filtered into 50 ml volumetric flask and kept for analysis. The potassium was determined with the help of ELICO flame photometer.

**Statistical Analysis:** LSD was used to compare all the means among treatments to compare the biochemical parameters.

## 3. RESULTS AND DISCUSSION

Biochemical basis of resistance was studied for 12 promising resistant and moderately resistant advanced backcross derived genotypes of Siddhi against BPH with regard to phenols, reducing sugars, ascorbic acid, nitrogen, phosphorous and potassium components in comparison with susceptible and resistant checks. The samples were taken before (healthy) and after release (infested) of BPH.

## 3.1 Phenols

Total phenolic content in the leaf sheaths of BPH infested and healthy rice plants was estimated in 12 advanced backcross derived genotypes of Siddhi along with the susceptible check TN1, resistant checks PTB33 and BM71. Total phenols in resistant checks PTB33 and BM71 increased from 0.217 to 0.256 mg g<sup>-1</sup> tissue (64.02%) and 0.204 to 0.232 mg g<sup>-1</sup> (62.3%), respectively. Whereas in the susceptible check (TN1) total phenols reduced from 0.186 to 0.133 mg g<sup>-1</sup> tissue of the leaf sheaths *i.e.*, by 28%. Significantly highest phenolic content (0.295 mg g<sup>-1</sup> tissue) was observed in infested Siddhi-BC2F6 BPHBL-61. The phenolic content increased in resistant genotypes after BPH infestation, the highest being 67.7% in Siddhi-BC2F6 BPH BL-64. The phenolic content in the healthy genotypes viz., Siddhi-BC2F6 BPH BL-11, Siddhi-BC2F6 BPH BL-19, Siddhi-BC2F6 BPH BL-24, Siddhi-BC2F6 BPH BL-56, Siddhi-BC2F6 BPH BL-57, Siddhi-BC2F6 BPH BL-60 and Siddhi-BC2F6 BPH BL-61 and the infested genotypes viz., Siddhi-BC2F6 BPH BL-57 and Siddhi-BC2F6 BPH BL-61 were significantly on par with resistant checks. Slight decrease in the phenolic content was observed in moderately resistant genotype Siddhi-BC2F6 BPH BL-19 and which was on par with susceptible check TN1 (Table 1).

The present results were in accordance to the findings of Reddy et al. [13] who reported that BPH infestation increased the phenolic content of the resistant and moderately resistant cultivars. The present results were similar with the findings of Udayasree et al. [14] who conducted studies

on biochemical aspects of resistance against BPH on resistant rice genotypes and stated that the amount of total phenol was observed to be maximum in the leaf sheath of moderately resistant variety. Similarly, Dharshini and Gowda [15] reported significant increase of phenol content as a result of BPH feeding. Deepa et al. [16] assessed the level of total phenols, crude silica and total sugars in BPH affected rice leaves, resistant varieties showed higher amount of phenols and crude silica as against low quantity of total sugars. Present results were in agreement with Singh [17] who reported that total phenol contents decreased in all infected plant parts.

## 3.2 Reducing Sugar

BPH infestation had greater influence on reducing sugar content. It was found that reducing sugar content was more in susceptible check than in resistant genotypes of Siddhi. The highest quantity of reducing sugar was present in susceptible check TN1 (1.211 mg g<sup>-1</sup> tissue) whereas in resistant checks PTB33 and BM71, reducing sugar content was decreased from 0.326 to 0.286 mg g<sup>-1</sup> tissue (12.4%) and 0.740 to 0.0.610 mg g<sup>-1</sup> (17.6%), respectively. The percentage reduction of reducing sugars in the resistant genotypes ranged from 18.7% to

36.2%. The reducing sugar content in the healthy genotypes *viz.*, Siddhi-BC2F6 BPH BL-11, Siddhi-BC2F6 BPH BL-12, Siddhi-BC2F6 BPH BL-19, Siddhi-BC2F6 BPH BL-52, Siddhi-BC2F6 BPH BL-56 and Siddhi-BC2F6 BPH BL-57 were significantly on par with resistant check BM71. The reducing sugar content in the infested genotypes *viz.*, Siddhi-BC2F6 BPH BL-11, Siddhi-BC2F6 BPH BL-12, Siddhi-BC2F6 BPH BL-19, Siddhi-BC2F6 BPH BL-52, Siddhi-BC2F6 BPH BL-56 and Siddhi-BC2F6 BPH BL-57 were significantly on par with resistant check BM71 (Table 2).

The present results were in agreement with the findings of Reddy et al. [13] who reported that the sugar content was reduced in TN1 after infestation. Similar results were reported by Udayasree et al. [14] who conducted studies on biochemical aspects of resistance to BPH on resistant rice genotype and stated that the amount of total sugars was lowest in RNR 26111 (0.33 mg g<sup>-1</sup>) and highest in susceptible check TN1 (2.97 mg g<sup>-1</sup>). The present results were similar to the findings of Jayasimha et al. [18] who reported that the soluble sugar content was reduced in the varieties after BPH damage. Similarly, Vanitha et al. [19] also reported that reducing sugar content was more in susceptible varieties than resistant varieties.

SI.	Paddy Genotypes	Phenols (mg g <sup>-1</sup> )				
No.		Healthy	Infested	% Increase		
1	Siddhi-BC2F6 BPH-BL-30(R)	0.150(0.387) <sup>f</sup>	0.191 (0.437) <sup>e</sup>	27.8		
2	Siddhi-BC2F6 BPH-BL-43(R)	0.138(0.371) <sup>f</sup>	0.198 (0.444) <sup>e</sup>	43.2		
3	Siddhi-BC2F6 BPH-BL-64(R)	0.148(0.384) <sup>f</sup>	0.248 (0.497) <sup>d</sup>	67.7		
4	Siddhi-BC2F6 BPH-BL 11(MR)	0.196(0.443) <sup>cd</sup>	0.282 (0.531) <sup>cd</sup>	43.7		
5	Siddhi-BC2F6 BPH-BL-12(MR)	0.159(0.399) <sup>ef</sup>	0.186 (0.431) <sup>ef</sup>	17.0		
6	Siddhi-BC2F6 BPH-BL-19(MR)	0.183(0.428) <sup>de</sup>	0.156 (0.395) <sup>fg</sup>	-14.7		
7	Siddhi-BC2F6 BPH-BL-24(MR)	0.183(0.425) <sup>de</sup>	0.260 (0.51) <sup>cd</sup>	42.0		
8	Siddhi-BC2F6 BPH-BL-52(MR)	0.163(0.403) <sup>ef</sup>	0.195 (0.441) <sup>e</sup>	19.9		
9	Siddhi-BC2F6 BPH-BL-56(MR)	0.201(0.448) <sup>cd</sup>	0.262 (0.512) <sup>cd</sup>	30.6		
10	Siddhi-BC2F6 BPH-BL-57(MR)	0.254(0.503) <sup>b</sup>	0.292 (0.54) <sup>bc</sup>	14.8		
11	Siddhi-BC2F6 BPH-BL-60(MR)	0.182(0.427) <sup>de</sup>	0.269 (0.519) <sup>cd</sup>	47.7		
12	Siddhi-BC2F6 BPH-BL-61(MR)	0.288(0.537) <sup>a</sup>	0.295 (0.543) <sup>bc</sup>	22.0		
13	TN1(S)	0.186(0.431) <sup>de</sup>	0.133 (0.364) <sup>g</sup>	-28.4		
14	PTB33(R)	0.217(0.466) <sup>cd</sup>	0.356 (0.596) <sup>a</sup>	64.1		
15	BM-71(R)	0.204(0.452) <sup>cd</sup>	0.332 (0.574) <sup>ab</sup>	62.3		
	CV	4.559	4.702			
	CD	0.033	0.041			

R= Resistant; MR= Moderately Resistant; S= Susceptible

Means in a column followed by same letter are not significantly different at 5% level byDMRT Figures in parenthesis are square root transformed values

SI.	Deddy Constynes	Reducing Sugars (mg g <sup>-1</sup> )			
No.	Paddy Genotypes	Healthy	Infested	% Decrease	
1	Siddhi-BC2F6 BPH-BL-30(R)	0.558 (0.746) <sup>def</sup>	0.398 (0.624) <sup>cde</sup>	28.6	
2	Siddhi-BC2F6 BPH-BL-43(R)	0.436 (0.659) <sup>fg</sup>	0.320 (0.547) <sup>def</sup>	26.6	
3	Siddhi-BC2F6 BPH-BL-64(R)	0.286 (0.535) <sup>hi</sup>	0.185 (0.424) <sup>f</sup>	35.4	
4	Siddhi-BC2F6 BPH-BL11(MR)	0.606 (0.775) <sup>cde</sup>	0.488 (0.698) <sup>bcd</sup>	19.5	
5	Siddhi-BC2F6 BPH-BL 12(MR)	0.775 (0.877) <sup>bc</sup>	0.630 (0.785) <sup>b</sup>	18.7	
6	Siddhi-BC2F6 BPH-BL-19(MR)	0.616 (0.784) <sup>cde</sup>	0.423 (0.639) <sup>bcde</sup>	31.3	
7	Siddhi-BC2F6 BPH-BL-24(MR)	0.420 (0.644) <sup>fgh</sup>	0.317 (0.557) <sup>def</sup>	24.6	
8	Siddhi-BC2F6 BPH-BL-52(MR)	0.702 (0.833) <sup>bcde</sup>	0.516 (0.717) <sup>bc</sup>	26.4	
9	Siddhi-BC2F6 BPH-BL-56(MR)	0.836 (0.914) <sup>b</sup>	0.534 (0.713) <sup>bc</sup>	36.2	
10	Siddhi-BC2F6 BPH-BL-57(MR)	0.631 (0.792) <sup>cde</sup>	0.480 (0.684) <sup>bcd</sup>	24.0	
11	Siddhi-BC2F6 BPH-BL-60(MR)	0.549 (0.74) <sup>ef</sup>	0.413 (0.636) <sup>bcde</sup>	24.7	
12	Siddhi-BC2F6 BPH-BL-61(MR)	0.271 (0.521) <sup>i</sup>	0.176 (0.416) <sup>f</sup>	35.0	
13	TN1(S)	1.211 (1.100) <sup>a</sup>	0.913 (0.955) <sup>a</sup>	24.6	
14	PTB33(R)	0.326 (0.567) <sup>ghi</sup>	0.286 (0.528) <sup>ef</sup>	12.4	
15	BM-71(R)	0.740 (0.859) <sup>bcd</sup>	0.61 (0.779) <sup>b</sup>	17.6	
	CV	3.006	3.154		
	CD	0.114	0.153		

## Table 2. Reducing sugars content of promising advanced backcross derived genotypes of Siddhi

R= Resistant; MR= Moderately resistant; S= Susceptible; Means in a column followed by same letter are not significantly different at 5% level byDMRT; Figures in parenthesis are square root transformed values

## Table 3. Ascorbic acid content of promising advanced backcross derived genotypes of Siddhi

SI.	Paddy Genotypes	Ascorbic Acid (mg g <sup>-1</sup> )			
No		Healthy	Infested	% Decrease	
1	Siddhi-BC2F6 BPH-BL-30(R)	1.153 (1.073) <sup>ab</sup>	0.899 (0.948) <sup>a</sup>	22.0	
2	Siddhi-BC2F6 BPH-BL-43(R)	0.557 (0.744) <sup>f</sup>	0.356 (0.589) <sup>ef</sup>	36.0	
3	Siddhi-BC2F6 BPH-BL-64(R)	1.225 (1.107) <sup>a</sup>	0.919 (0.958) <sup>a</sup>	25.0	
4	Siddhi-BC2F6 BPH-BL-11(MR)	0.769 (0.876) <sup>cd</sup>	0.646 (0.803) <sup>b</sup>	16.0	
5	Siddhi-BC2F6 BPH-BL-12(MR)	0.563 (0.749) <sup>ef</sup>	0.461 (0.676) <sup>cde</sup>	18.1	
6	Siddhi-BC2F6 BPH-BL-19(MR)	0.757 (0.869) <sup>cd</sup>	0.625 (0.79) <sup>bc</sup>	17.4	
7	Siddhi-BC2F6 BPH-BL-24(MR)	0.820 (0.903) <sup>cd</sup>	0.674 (0.82) <sup>b</sup>	17.8	
8	Siddhi-BC2F6 BPH-BL-52(MR)	0.554 (0.743) <sup>f</sup>	0.416 (0.643) <sup>de</sup>	24.9	
9	Siddhi-BC2F6 BPH-BL-56(MR)	0.760 (0.864) <sup>cde</sup>	0.568 (0.73) <sup>bcd</sup>	25.3	
10	Siddhi-BC2F6 BPH-BL-57(MR)	0.703 (0.833) <sup>def</sup>	0.508 (0.711) <sup>bcd</sup>	27.8	
11	Siddhi-BC2F6 BPH-BL-60(MR)	0.755 (0.868) <sup>cd</sup>	0.617 (0.785) <sup>bc</sup>	18.3	
12	Siddhi-BC2F6 BPH-BL-61(MR)	0.930 (0.964) <sup>ab</sup>	0.665 (0.812) <sup>b</sup>	28.5	
13	TN1(S)	0.632 (0.794) <sup>def</sup>	0.258 (0.508) <sup>f</sup>	59.1	
14	PTB33(R)	1.118 (1.056) <sup>ab</sup>	0.959 (0.979) <sup>a</sup>	14.2	
15	BM-71(R)	1.043 (1.021) <sup>ab</sup>	0.918 (0.958) <sup>a</sup>	12.0	
	CV	3.762	3.892		
	CD	0.117	0.116		

R= Resistant; MR= Moderately resistant; S= Susceptible; Means in a column followed by same letter are not significantly different at 5% level by DMRT; Figures in parenthesis are square root transformed values

## 3.3 Ascorbic Acid

The studies on ascorbic acid content of leaf sheath of resistant genotypes indicated that the

quantity of ascorbic acid in healthy genotypes of Siddhi-BC2F6 BPH BL-64, Siddhi-BC2F6 BPH BL-30 and Siddhi-BC2F6 BPH BL-61 were significantly on par with the resistant checks. The ascorbic acid content in susceptible check TN1 was found as 0.632 mg g<sup>-1</sup>. Whereas in resistant checks PTB33 and BM71, the percentage decrease in ascorbic acid content was 14.2% and 12%, respectively. The highest quantity of ascorbic acid was present in Siddhi-BC2F6 BPH BL-64 (1.225 mg g<sup>-1</sup>). The ascorbic content in infested genotypes viz., Siddhi-BC2F6 BPH BL-30 and Siddhi-BC2F6 BPH BL-64 were significantly on par with resistant checks. The highest percentage decrease observed in susceptible check TN1 (59.1%). Percentage reduction of ascorbic acid content in the infested genotypes ranged from 16% to 36% (Table 3). The results obtained were similar with the findings of Ramulamma et al. [20] who reported that the BPH infestation caused decrease in ascorbic acid content in infested rice cultures.

## 3.4 Nitrogen

The nitrogen (N) content in the backcross derived genotypes, resistant check (PTB33) and susceptible check (TN1) varied. The highest percentage of nitrogen (0.91%) was found in the healthy susceptible check TN1 while in the resistant and moderately resistant genotypes, the total N content ranged from 0.73% to 0.89%, thus showed significantly lower N content than susceptible check TN1. The total N content in the genotypes i.e., Siddhi-BC2F6 BPH BL-43 and Siddhi-BC2F6 BPH BL-24 was significantly on par with the resistant checks. In general, the total N content in the infested genotypes decreased over healthy plants in most of the genotypes. The percentage reduction in N content of the ranged from 6.8% to genotypes 25.3%. Percentage decrease in the N content was highest in the susceptible check TN1 (35.2%). Whereas in the resistant checks, PTB33 and BM71 decrease in the N content was 10.1% and 12.5%, respectively (Table 4).

The present results were in accordance with the findings of Dharshini and Gowda [15] who reported after BPH infestation, the total nitrogen was reduced in all varieties after BPH damage. The present results were similar with the findings of Watanabe and Kitigawa [21] who reported that BPH feeding on rice plants caused decrease in the contents of N and thus reduced the growth of main shoot and tillers.

## 3.5 Phosphorus

The percentage increase of phosphorous content in the resistant genotypes ranged from 4.6% to

12.9%. Whereas in resistant checks PTB33 and BM71, the percentage increase in phosphorous content was 11.1% and 15%, respectively. The phosphorous content in healthy genotypes Siddhi-BC2F6 BPH BL-12, Siddhi-BC2F6 BPH BL-19, Siddhi-BC2F6 BPH BL- 56, Siddhi-BC2F6 BPH BL-57 and Siddhi-BC2F6 BPH BL-61 was significantly on par with the resistant checks. However, in Siddhi-BC2F6 BPH BL-11 (2.3%) decrease in the phosphorus content was recorded (Table 5). The results obtained were inconformity with findings of Ramulamma et al. [20] who reported marginal differences in the phosphorus content of the infested cultures compared to uninfested cultures. Similar results were reported by Vanitha et al. [19] who stated that the per cent reduction of P and K was much less upon BPH infestation compared to N content.

## 3.6 Potassium

Percentage increase of potassium content in resistant genotypes ranged from 7.4% to 33.1%. The potassium content in resistant checks PTB33 and BM71 increased from 16% and 14.1%, respectively. Whereas the resistant and moderately resistant genotypes showed significant increase in the potassium content after BPH infestation. The potassium content in the healthy resistant genotypes *i.e.*, Siddhi-BC2F6 BPH BL-30, Siddhi-BC2F6 BPH BL-43, Siddhi-BC2F6 BPH BL-11, Siddhi-BC2F6 BPH BL-24, Siddhi-BC2F6 BPH BL-56, Siddhi-BC2F6 BPH BL-57, Siddhi-BC2F6 BPH BL-60 was significantly on par with the resistant checks. Significantly highest potassium content (0.274%) infested Siddhi-BC2F6 was recorded in compared **BPHBL-64** to other infested backcross derived genotypes. Potassium content in all the resistant genotypes recorded an increase after BPH infestation, the highest being 33.1% in Siddhi-BC2F6 BPH BL-61 (Table 6).

The present results were in conformity with the findings of Ramulamma et al. [20] who recorded the significant increase in the potassium content in the resistant and moderately resistant cultures of rice after infestation. Similarly, Vanitha et al. [19] reported that K content was slightly increased in the BPH infested plants. Liu et al. [22] also reported that BPH infestation caused a reduction in the K uptake of roots in rice varieties and the influence became more serious with the increase of BPH density and the prolongation of infestation

duration. From the present study it can be concluded that the infestation of BPH leads to reduction in nitrogen, ascorbic acid and reducing sugars content but increase in phenols, phosphorus and potassium contents, compared to the susceptible check the increase is greater in resistant and moderately resistant genotypes [23-25].

SI.	Paddy Genotypes	Nitrogen (%)			
No		Healthy	Infested	% Decrease	
1	Siddhi-BC2F6 BPH-BL-30(R)	0.84(1.16) <sup>cde</sup>	0.75(1.12) <sup>bc</sup>	10.7	
2	Siddhi-BC2F6 BPH-BL-43(R)	0.76(1.12) <sup>ghi</sup>	0.67(1.08) <sup>efg</sup>	11.8	
3	Siddhi-BC2F6 BPH-BL-64(R)	0.86(1.17) <sup>cd</sup>	0.79(1.14) <sup>ab</sup>	8.10	
4	Siddhi-BC2F6 BPH-BL-11(MR)	0.89(1.18) <sup>ab</sup>	0.80(1.14) <sup>a</sup>	10.1	
5	Siddhi-BC2F6 BPH-BL-12(MR)	0.73(1.11) <sup>i</sup>	0.68(1.09) <sup>ef</sup>	6.80	
6	Siddhi-BC2F6 BPH-BL-19(MR)	0.81(1.14) <sup>ef</sup>	0.66(1.08) <sup>efg</sup>	18.5	
7	Siddhi-BC2F6 BPH-BL-24(MR)	0.79(1.13) <sup>gh</sup>	0.62(1.06) <sup>hi</sup>	21.5	
8	Siddhi-BC2F6 BPH-BL-52(MR)	0.84(1.16) <sup>bc</sup>	0.77(1.13) <sup>bc</sup>	8.30	
9	Siddhi-BC2F6 BPH-BL-56(MR)	0.81(1.14) <sup>ef</sup>	0.73(1.11) <sup>cd</sup>	9.90	
10	Siddhi-BC2F6 BPH-BL-57(MR)	0.87(1.17) <sup>bc</sup>	0.65(1.07) <sup>gh</sup>	25.3	
11	Siddhi-BC2F6 BPH-BL-60(MR)	0.88(1.17) <sup>bc</sup>	0.75(1.12) <sup>bc</sup>	14.8	
12	Siddhi-BC2F6 BPH-BL-61(MR)	0.85(1.16) <sup>cde</sup>	0.64(1.07) <sup>gh</sup>	24.7	
13	TN1(S)	0.91(1.19) <sup>a</sup>	0.59(1.04) <sup>i</sup>	35.2	
14	PTB33(R)	0.79(1.14) <sup>gh</sup>	0.71(1.10) <sup>cde</sup>	10.1	
15	BM-71(R)	0.80(1.14) <sup>g</sup>	0.70(1.10) <sup>cde</sup>	12.5	
	CV	4.07	4.27		
	CD	0.062	0.058		

R= Resistant; MR= Moderately resistant; S= Susceptible; Means in a column followed by same letter are not significantly different at 5% level by DMRT; Figures in parenthesis are square root transformed values

SI.	Paddy Genotypes	Phosphorous (%)				
No.		Healthy	Infested	% Increase		
1	Siddhi-BC2F6 BPH-BL-30(R)	0.519 (0.72) <sup>b</sup>	0.557 (0.746) <sup>bcd</sup>	7.40		
2	Siddhi-BC2F6 BPH-BL-43(R)	0.613 (0.783)ª	0.675 (0.821)ª	10.1		
3	Siddhi-BC2F6 BPH-BL-64(R)	0.635 (0.797) <sup>a</sup>	0.664 (0.815) <sup>a</sup>	4.60		
4	Siddhi-BC2F6 BPH-BL-11(MR)	0.617 (0.785)ª	0.603 (0.776) <sup>abc</sup>	-2.30		
5	Siddhi-BC2F6 BPH-BL-12(MR)	0.461 (0.679) <sup>c</sup>	0.513 (0.716) <sup>d</sup>	11.2		
6	Siddhi-BC2F6 BPH-BL-19(MR)	0.495 (0.703) <sup>bc</sup>	0.540 (0.734) <sup>cd</sup>	9.10		
7	Siddhi-BC2F6 BPH-BL-24(MR)	0.382 (0.618) <sup>d</sup>	0.505 (0.709) <sup>d</sup>	32.0		
8	Siddhi-BC2F6 BPH-BL-52(MR)	0.590 (0.768) <sup>a</sup>	0.638 (0.798) <sup>ab</sup>	8.10		
9	Siddhi-BC2F6 BPH-BL-56(MR)	0.479 (0.692) <sup>bc</sup>	0.519 (0.72) <sup>cd</sup>	8.40		
10	Siddhi-BC2F6 BPH-BL-57(MR)	0.469 (0.685) <sup>bc</sup>	0.529 (0.727) <sup>cd</sup>	12.9		
11	Siddhi-BC2F6 BPH-BL-60(MR)	0.595 (0.771) <sup>a</sup>	0.652 (0.807) <sup>a</sup>	9.60		
12	Siddhi-BC2F6 BPH-BL-61(MR)	0.368 (0.606) <sup>de</sup>	0.413 (0.641) <sup>e</sup>	12.2		
13	TN1	0.481 (0.693) <sup>bc</sup>	0.521 (0.721) <sup>cd</sup>	8.20		
14	PTB33	0.325 (0.57) <sup>e</sup>	0.361 (0.601) <sup>e</sup>	11.1		
15	BM-71	0.461 (0.678) <sup>c</sup>	0.53 (0.726) <sup>cd</sup>	15.0		
	CV	3.344	4.825			
	CD	0.039	0.059			

	Table 5. Ph	osphorous	content of	promising	advanced	backcross	derived	genotypes	ofSiddhi
--	-------------	-----------	------------	-----------	----------	-----------	---------	-----------	----------

R= Resistant; MR= Moderately resistant; S= Susceptible; Means in a column followed by same letter are not significantly different at 5% level byDMRT; Figures in parenthesis are square root transformed values

SI.	Paddy Genotypes	Potassium (%)				
No.		Healthy	Infested	% Increase		
1	Siddhi-BC2F6 BPH-BL-30(R)	0.190 (0.436) <sup>cdef</sup>	0.222 (0.471) <sup>e</sup>	16.8		
2	Siddhi-BC2F6 BPH-BL-43(R)	0.232 (0.481) <sup>b</sup>	0.279 (0.528) <sup>abc</sup>	20.1		
3	Siddhi-BC2F6 BPH-BL-64(R)	0.274 (0.524) <sup>a</sup>	0.297 (0.548)ª	8.30		
4	Siddhi-BC2F6 BPH-BL-11(MR)	0.224 (0.473) <sup>bc</sup>	0.283 (0.542) <sup>ab</sup>	26.5		
5	Siddhi-BC2F6 BPH-BL-12(MR)	0.159 (0.399) <sup>fg</sup>	0.212 (0.473) <sup>e</sup>	33.1		
6	Siddhi-BC2F6 BPH-BL-19(MR)	0.129 (0.359) <sup>h</sup>	0.164 (0.404) <sup>f</sup>	26.9		
7	Siddhi-BC2F6 BPH-BL-24(MR)	0.208 (0.455) <sup>bcd</sup>	0.233 (0.488) <sup>cde</sup>	12.2		
8	Siddhi-BC2F6 BPH-BL-52(MR)	0.171 (0.414) <sup>efg</sup>	0.158 (0.4) <sup>f</sup>	-8.00		
9	Siddhi-BC2F6 BPH-BL-56(MR)	0.217 (0.464) <sup>bc</sup>	0.233 (0.485) <sup>de</sup>	7.40		
10	Siddhi-BC2F6 BPH-BL-57(MR)	0.198 (0.443) <sup>bcde</sup>	0.217 (0.476) <sup>e</sup>	9.80		
11	Siddhi-BC2F6 BPH-BL-60(MR)	0.216 (0.464) <sup>bc</sup>	0.256 (0.519) <sup>abcd</sup>	18.5		
12	Siddhi-BC2F6 BPH-BL-61(MR)	0.179 (0.423) <sup>def</sup>	0.236 (0.486) <sup>de</sup>	31.8		
13	TN1	0.144 (0.38) <sup>gh</sup>	0.153 (0.391) <sup>f</sup>	5.80		
14	PTB33	0.225 (0.474) <sup>bc</sup>	0.261 (0.506) <sup>cde</sup>	16.0		
15	BM-71	0.224 (0.472) <sup>bc</sup>	0.256 (0.502) <sup>bcde</sup>	14.1		
	CV	3.386	4.261			
	CD	0.04	0.042			

Table 6. Potassium content of promising advanced backcross derived genotypes ofSiddhi

*R*= Resistant; *M*R= Moderately Resistant; *S*= Susceptible; Means in a column followed by same letter are not significantly different at 5% level byDMRT; Figures in parenthesis are square root transformed values

#### 4. CONCLUSION

The BPH infestation in resistant genotypes and resistant checks resulted in an increase in the phenolic content in the infested plant, whereas in the susceptible TN1, reduction was observed. Significantly highest phenolic content (0.295 mg g-1 tissue) was observed in infested Siddhi-BC2F6 BPH-BL-61 compared to all genotypes. The total reducing sugar content was significantly highest in the healthy susceptible check TN1 (1.211 mg g-1). There was a decrease in total reducing sugars in all the plants upon BPH infestation. In infested genotypes there was a reduction in the ascorbic acid content in almost all the genotypes including the checks. The lowest quantity of ascorbic acid in healthy resistant and moderately resistant genotypes present in Siddhi-BC2F6 BPH BL-52, Siddhi-BC2F6 BPH BL-43 and Siddhi-BC2F6 BPH BL-12 (0.554 mg g  $^{-1}$  , 0.557 mg g  $^{-1}$  and 0.563 mg g  $^{-1},$ respectively) which were on par with the susceptible check TN1 (0.632 mg g<sup>-1</sup>) while the highest ascorbic acid quantity was present in Siddhi-BC2F6 BPH BL-30 and Siddhi-BC2F6 BPH BL-64 (1.153 mg g<sup>-1</sup> and 1.225 mg g<sup>-1</sup>). The healthy plants of susceptible TN1 had higher nitrogen content. Significant decrease of nitrogen was recorded in infested plants of susceptible TN1 (35.2%) than the resistant genotypes. The percentage decrease in the nitrogen % of the genotypes ranged from 8.1% to 25.3%. There

was increase in phosphorus content in resistant and moderately resistant genotypes before and after infestation, the highest phosphorous % was recorded in infested Siddhi-BC2F6 BPH-BL-43 (0.675%). The highest potassium % was present in infested Siddhi-BC2F6 BPH-BL-64 (0.297%) followed by Siddhi-BC2F6 BPH-BL-11 (0.283%). Increase in potassium content was observed in resistant and moderately resistant genotypes and slight increase was recorded in susceptible check TN1 after BPH infestation. Overall, it can be concluded that upon BPH infestation in resistant and moderately resistant genotypes higher amounts of phenols, potassium, phosphorous and reduced level of nitrogen. reducing sugars and ascorbic acid were observed.

#### **5. FUTURE SCOPE**

Understanding the biochemical pathways and defense mechanisms can pave the way for identifying key resistance genes and biochemical markers, facilitating marker-assisted selection in breeding programs.

#### DISCLAIMER (ARTIFICIAL INTELLIGENCE)

Author(s) hereby declare that NO generative AI technologies such as Large Language Models (ChatGPT, COPILOT, etc) and text-to-image

generators have been used during writing or editing of manuscripts.

## **COMPETING INTERESTS**

Authors have declared that no competing interests exist.

## REFERENCES

- Molla KA, Karmakar S, Molla J, Bajaj P, Varshney RK, Datta SK, Datta K. Understanding sheath blight resistance in rice: The road behind and the road ahead. Plant Biotechnology Journal. 2020; 18(4):895-915.
- Food and agricultural organization. FAO STAT database FAO; 2023. Available:WWW.Faostat.Org.
- 3. USDA-International Production Assessment Division; 2023. Available:https://ipad.fas.usda.gov/cropexp lorer/util/new\_get\_psd\_data.aspx?regio nid=sasia
- Bhogadhi SC, Bentur JS, Rani CVD, Thappeta G, Yamini KN, Kumar NAP, Satynarayana PV. Screening of rice genotypes for resistance to brown plant hopper biotype 4 and detection of BPH resistance genes. International Journal of Life Sciences Biotechnology and Pharma Research. 2015;4(2):90.
- 5. Atwal A, Dhaliwal G. Agricultural Pest of South–East Asia & Their Management; 2002.
- Heong KL, Hardy B. (Eds.). Planthoppers: New threats to the sustainability of intensive rice production systems in Asia. Int. Rice Res. Inst; 2009.
- Vanitha K, Suresh S, Gunathilagaraj K. Influence of brown planthopper *Nilaparvata lugens* (Stal.) feeding on nutritional biochemistry of rice plant. *Oryza*. 2021;48 (2):2249-5266.
- 8. Kumari RP, Anusha C, Kumar RS, Prasad KR, Hari Y. Screening of Rice Genotypes for their Resistance against Brown Plant Hopper, *Nilaparvata lugens* (Stal) under Field and Glasshouse Conditions; 2022.
- Nelson N. A photometric adaptation of the Somogyi method for the determination of glucose. J. biol. Chem. 1944;153(2):375-380.
- 10. Sadasivan & Manickam. Biochemical methods, second edition. New Age

International Publisher (P) Limited, New Delhi; 1966.

- Ma T, Zuazaga G. Micro-Kjeldahl determination of nitrogen. A new indicator and an improved rapid method. Industrial & Engineering Chemistry Analytical Edition. 1942;14(3):280-282.
- 12. Piper CS. Soil and Plant Analysis. Scientific Publishers; 2019.
- Reddy KL, Pasalu IC, Raju AS, Reddy DDR. Biochemical basis of resistance in rice cultivars to brown plant hopper *Nilaparvata lugens* (Stal.). Journal of Entomological Research. 2004;28(1):79-85.
- Udayasree M, Rajanikanth P, Varma NRG, Sreedhar M. Studies on Non-preference mechanism and biochemical aspects of resistance to BPH *Nilaparvata lugens* (Stal.) on resistant rice genotypes. Methodology; 2018.
- 15. Dharshini GM, Gowda DS. Biochemical basis of resistance in rice landraces to brown planthopper, *Nilaparvata lugens* (Stal.); 2014.
- Deepa K, Pillai MA, Murugesan N. Biochemical bases of resistance to brown planthopper (*Nilaparvata lugens*) (Stål) in different rice accessions. Agricultural Science Digest-A Research Journal. 2016; 36(2):102-105.
- 17. Singh HV. Biochemical transformation in *Brassica* spp. due to *Peronospora parasitica* infection. Annals of Plant Protection Sciences. 2004;12(2):292 -295.
- Jayasimha GT, Nalini R, Chinniah C, Muthamilan M, Mini ML. Evaluation of biochemical constituents in healthy and brown planthopper, *Nilaparvata lugens* (Stal.) (Hemiptera: Delphacidae) Damaged Rice Plants; 2015.
- Vanitha K, Suresh S, Gunathilagaraj K. Influence of brown planthopper *Nilaparvata lugens* (Stal.) feeding on nutritional biochemistry of rice plant. ORYZA-An International Journal on Rice. 2011;48(2): 142-146.
- 20. Ramulamma A, Sridevi D, Jhansilakshmi V, Reddy BN. Biochemical analysis of resistance in paddy to BPH, *Nilaparvata lugens*. Environment & Ecology. 2017; 26:6.
- 21. Watanabe T, Kitagawa H. Photosynthesis and translocation of assimilates in rice plants following phloem feeding by the planthopper *Nilaparvata lugens*

(Homoptera: Delphacidae). Journal of Economic Entomology. 2000;93(4):1192-1198.

- 22. Lu ZX, Heong KL, Yu XP, Hu C. Effects of nitrogen on the tolerance of brown planthopper, *Nilaparvata Lugens*, to adverse environmental factors. Insect Science. 2005;12(2):121-128.
- VI S. Analysis of total phenols and other oxidation substrates and antioxidants by means of Folin-Ciocalteu reagent. Methods in Enzymology. 1999; 299:152-178.
- 24. Pati P, Jena M, Bhattacharya S, Behera SK, Pal S, Shivappa R, Dhar T. Biochemical defense responses in red rice

genotypes possessing differential resistance to brown planthopper, Nilaparvata lugens (Stål). Insects. 2023;14 (7):632. Available:https://www.mdpi.com/2075-

4450/14/7/632

25. Senthil-Nathan Kalaivani K, S. Choi MY. Paik CH. Effects of iasmonic acid-induced resistance the in rice on plant brownhopper, Nilaparvata lugens Stål (Homoptera: Delphacidae). Pesticide Biochemistry and Physiology. 2009;95(2): 77-84.

Available:https://www.sciencedirect.com/sc ience/article/pii/S0048357509000790

**Disclaimer/Publisher's Note:** The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of the publisher and/or the editor(s). This publisher and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.

© Copyright (2024): Author(s). The licensee is the journal publisher. This is an Open Access article distributed under the terms of the Creative Commons Attribution License (http://creativecommons.org/licenses/by/4.0), which permits unrestricted use, distribution, and reproduction in any medium, provided the original work is properly cited.

Peer-review history: The peer review history for this paper can be accessed here: https://www.sdiarticle5.com/review-history/120142