



## Biochemical Characterization of Brinjal (*Solanum melongena* L.) Leaves Infected with Little Leaf Disease

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### Abstract

The experiment was conducted during September, 2023 to February, 2024 at the Department of Biochemistry, Anand Agricultural University, Anand, Gujarat, India. Total 28 genotype originating from a cross between a little leaf resistance parent AB 15-06 (*S. melongena*) and a susceptible parent GRB 5 (*S. melongena*). Disease incidence recorded from 14 each of highly resistant and susceptible genotypes and their parents AB 15-06 (Resistance against little leaf disease) and GRB 5 (Susceptible against little leaf disease) were selected for biochemical attributes comprised of chlorophyll (0.73–0.98 mg g<sup>-1</sup>), moisture (76.77–97.43%), phenol (0.43–3.26 mg g<sup>-1</sup>), total soluble sugar (2.36–4.57 mg g<sup>-1</sup>), membrane injury (24.56–69.87%). Correlation analysis of biochemical characters like chlorophyll and moisture were observed negatively correlated with disease incidence while rest of biochemical parameters total soluble solids, membrane injury and phenol content were positively correlated. Membrane injury (0.254), moisture (0.026), TSS (0.030), and phenol content (0.016) showed positive skewness whereas chlorophyll (-1.318), membrane injury (-1.540), moisture (-1.543), total soluble sugar (-1.160) and phenol content (-1.626) detected negative kurtosis. GCV, PCV and heritability analysis for all biochemical characters recorded were chlorophyll (36.46%, 36.5% and 99.82%), membrane injury (61.52%, 61.96% and 98.59%), moisture (7.55%, 8.73% and 13.44%), total soluble sugar (99.39%) and phenol content (46.76%, 46.83% and 99.67%).

**Keywords:** Brinjal, correlation, GCV, little leaf disease, PCV

### 1. Introduction

Brinjal (*Solanum melongena* L.) is a widely produced vegetable crop in tropical and subtropical regions that is valued for its tasty fruit, which is categorized as a berry by nature. It is commonly referred to as “aubergine” in Europe, “eggplant” in the Americas and Australia, and “brinjal” in South Asia, South-east Asia, and South Africa. Brinjal is considered the poor man’s vegetable because to its tremendous productivity. This particular food type comes with inherent medicinal values compounded by the fact that it tastes good and is healthy to the body (Naeem and Ugur, 2019). Brinjal is a fairly good source of iron, calcium, phosphorus, potassium and vitamin B group. It’s fresh weight is composed of 92.7% moisture, 1.4% protein, 1.3% fibre, 0.3% fat, 0.3% minerals and remaining 4% consist of carbohydrates and vitamins (A and C). Brinjal is rich source of secondary metabolites, antioxidant, polyphenols, vitamins which is useful for combating pathogen against disease (Oladosu et al., 2021, Sharma and kaushik, 2021). White kinds of brinjal fruits, leaves and root are very helpful

to the diabetic patients and contain a good healer property which is used for many ayurvedic medicines (Yarmohammadi et al., 2021, Colak et al., 2022). Large quantities of anthocyanin and phenolic component which is present in brinjal that acts as the most significant antioxidant and is involved in numerous physiological activities like anticancer properties, antimutagenics and vision improvement (Gürbüz et al., 2018, Tena et al., 2020, Condurache et al., 2021).

Brinjal is afflicted at various stages by a variety of diseases, which results in significant output losses. In India, *Datura stramonium* was identified as a naturally occurring weed host for BLL phytoplasma *Hishimonas phycitis*, a leafhopper, was found to be a possible vector (Rao et al., 2021, Karthikeyan et al., 2024). The insect vector of little leaf disease is *Hishimonus phycitis*, a member of the leafhopper family. It severely reduces this crop’s output. During the disease certain changes occur in crops physiological and biochemical condition. The afflicted plants exhibit small leaves, an abundance of new shoots, phyllody, and stunting (Kumar et al., 2017, Kumari et



al., 2019, Dutta et al., 2022). Numerous yellow-type diseases were earlier thought to be caused by viruses in view of their infective spread, their symptomatology and the fact that they were transmitted by insects. This yellow diseases causing microorganism were first time observed by Japanese workers as pleomorphic cells in ultra-thin sections of leaves of mulberry infected with dwarf disease (Wang et al., 2024). Phytoplasma are round to filamentous, 200–800 nm diameter, gram positive, insect-transmitted, plant phloem limited and non culturable prokaryotes, smallest among known cellular plant parasites and pathogens which are self replicating by transverse binary fission that can cause devastating loss in crops and natural ecosystem worldwide (Bertaccini, 2022). They are morphologically more resembled to the mycoplasma hence called as a mycoplasma like organisms (MLO's) earlier, but after the introduction of molecular technologies it was realized that this wall less plant infecting prokaryotes are different from the mycoplasma in many characteristics than so the trivial name "phytoplasma" was adapted in 10th congress of International Organization of Mycoplasma in 1994. Phytoplasma genetically more resemble to the acheloplasma (Namba, 2019). This study main aim to characterized different biochemical parameter which affected during little leaf disease infection and comparison with healthy and disease infected brinjal leaves which is further used to identification of trait association of biochemical markers in brinjal genotypes.

## 2. Materials and Methods

### 2.1. Experimental material

The experimental material for present investigation comprised total 28 genotype originating from a cross between a little leaf resistance parent of brinjal AB 15-06 (*S. melongena*) and a susceptible parent GRB 5 (*S. melongena*) in year 2023–2024 in month of September to February at Main Vegetable Research Station, Anand Agricultural University, Anand, Gujarat, India. From these total 28 genotypes, 14 healthy and 14 little leaf infected were selected for analysis of biochemical parameter like chlorophyll, moisture, total soluble sugar, phenols and membrane injury. Lab experiment was carried out at Department of Biochemistry, Anand Agricultural University, Anand, Gujarat, India.

### 2.2. Biochemical characterization of brinjal genotypes and their parents

#### 2.2.1. Chlorophyll

Hiscox and Israelstam (1979) method was used to determined the amount of chlorophyll present in brinjal leaves. After adding 10 ml of DMSO (dimethyl sulfoxide) containing 50 mg of fresh leaves in test tubes, the mixture was incubated at 12 hrs at 27°C. DMSO was used as a blank without leaves sample. The change in color was recorded at wavelengths 663 nm and 645 nm in spectrophotometer and chlorophyll a, b and total chlorophyll was quantified as per the given formula.

Chlorophyll a ( $\text{mg g}^{-1} \text{ f.w.}$ ) =  $(12.7 \times \text{O.D. at } 663 \text{ nm} - 2.69 \times \text{O.D. at } 645 \text{ nm}) / (1000 \times 0.05)$  .....(1)

Chlorophyll b ( $\text{mg g}^{-1} \text{ f.w.}$ ) =  $(22.7 \times \text{O.D. at } 645 \text{ nm} - 4.68 \times \text{O.D. at } 663 \text{ nm}) / (1000 \times 0.05)$  .....(2)

Total chlorophyll ( $\text{mg g}^{-1} \text{ f.w.}$ ) = Chlorophyll a + Chlorophyll b .....(3)

#### 2.2.2. Moisture

The moisture content was estimated as per the procedure developed by Sadasivam and Manickam (1992). Five grams of brinjal leaves sample were raped in the small aluminum foil boxes. The weight of each box with leaves was pre-recorded then the aluminum foil box with containing samples were kept in the hot air oven at 105°C for 6 hrs. All boxes were taken out of the oven and allowed to cool at room temperature in desiccator. The amount of total moisture was calculated as per following formula,

Moisture (%) =  $(\text{Fresh weight (g)} - \text{Dry weight (g)}) / (\text{Fresh weight (g)}) \times 100$  .....(4)

#### 2.2.3. Total phenol

Total phenol was estimated as a method described by Dhruv et al. (2021) One gram of brinjal leaves was homogenized in 80% methanol using mortar and pestle and the final volume was made to 10 ml. The content was refluxed for two hours on boiling water bath at 65°C. Supernatant was collected and the residue was re-extracted twice with 80% methanol. All supernatants were combined, and the final volume was made to 10 ml. The extract was used for the assay of total phenol. Aliquot 0.2 ml was taken and made the final volume 1.0 ml with distilled water. For standard, catechol (50 to 250 microgram working standard in water) was added in separate test tubes. 0.2, 0.4, 0.6, 0.8, and 1 ml. To this add 1 ml of folin reagent and after 3 min 2 ml of 20%  $\text{Na}_2\text{CO}_3$  was added, and the tubes were incubated at room temperature for 30 min and made the total volume of 5 ml with distilled water. The absorbance was measured at 650 nm. Phenol content was calculated using following formula.

Phenol (%) =  $(\text{Reading} \times \text{Graph factor} \times \text{Total volume} \times 10^{-6}) / (\text{Taken volume} \times \text{Sample weight (g)})$  .....(5)

#### 2.2.4. Total soluble sugar

Total soluble sugars were determined using the phenol-sulphuric acid method as described by Dubois et al. (1956) with some modification. Total Soluble sugar was extracted from 0.5 g of brinjal leaves in 5 ml 80% methanol. Which was then incubated for 2 hr in a shaker. After incubation centrifugation was done and clear supernatant was collected in another test tube. Supernatant was evaporated in boiling water bath. To the residue 1 ml of distil water was added and used to quantified sugar content. For stock solution 500 mg of reagent grade glucose in a chemical balance and dissolved in 100 ml of distilled water. Then 1 ml of aliquot and diluted further to 100 ml which gave  $50 \mu\text{g ml}^{-1}$ , from this 0.5, 1.0, 1.5, 2.0, and 2.5 ml of the working standard glucose solution was used for standard curve preparation in the range of 25 to 125  $\mu\text{g}$ . Makeup the final volume of 3 ml with the addition

of distilled water was pipetted into a series of tubes. Add 5% of 1 ml phenol in all test tubes. After that incubate for 3 min at room temperature. To this, 5.0 ml concentrated sulphuric acid was carefully added to the side of the tube. After mixing thoroughly the tubes were kept for 10 m at room temperature and 20 min in a cold-water bath for color development. The absorbance was measured at 490 nm. Total soluble sugar was calculated by using given formula.

Total soluble sugars (%) =  $\frac{\text{Reading} \times \text{Graph factor} \times \text{Total volume} \times 10^{-6}}{\text{Taken volume} \times \text{Sample weight (g)}}$  .....(6)

#### 2.2.5. Membrane injury

Membrane injury was determined by using the method of Sairam et al. (2002) with some modifications. For determination of membrane injury of brinjal genotypes, fresh leaves tissue i.e. 0.1 g were taken. These tissues were placed in test tubes containing 100 ml of distilled water. They were kept in hot water bath at 40°C for 30 m. After 30 m samples were cooled to room temperature and electrical conductivity (EC) were recorded for both control and treatment ( $C_1$  and  $T_1$ ). Then the tubes were kept in boiling water bath at 100°C for 15 m. After cooling, again electrical conductivity (EC) were measured ( $C_2$  and  $T_2$ ).

Membrane injury was calculated by using the following formula:

$$MI\% = 1 - \frac{(1 - T_1/T_2)}{(1 - C_1/C_2)} \times 100 \quad \dots\dots\dots(7)$$

Where,  $T_1$  – Electric conductivity of Disease leaves at 40°C for 30 m

$T_2$  – Electric conductivity of Disease leaves at 100°C for 15 m

$C_1$  – Electric conductivity of Healthy leaves at 40°C for 30 m

$C_2$  – Electric conductivity of Healthy leaves at 100°C for 15 m

#### 2.3. Disease incidence of little leaf infection in brinjal genotypes

Random observations of little leaf disease scoring in brinjal was carried out at weekly interval after 30 days of transplanting. Little leaf disease incidence data were recorded from selected genotypes and their parents AB 15-06 and GRN 5. Based on % disease incidence, the brinjal genotypes were classified into five categories (Venkataravanappa et al., 2022).

1. Immune (0%)
2. Resistant (0.1–10%)
3. Moderately resistant (10.1–20%)
4. Susceptible (20.1–50%), and
5. Highly susceptible (>50%)

#### 2.4. Statistical analysis

##### 2.4.1. Analysis of variance

To assess the variations in brinjal genotypes for all biochemical characters, the Panse and Sukhtme (1967) analysis of variance technique was employed.

##### 2.4.2. Phenotypic (PCV) and genotypic (GCV) coefficients of variations

Phenotypic and genotypic co-efficient of variation was calculated by method described by Burton and Devane (1953).

##### 2.4.3. Genotypic coefficient of variation (GCV %)

Genotypic coefficient of variation was calculated using the following formula given below.

$$GCV\% = \frac{\sqrt{\sigma^2_g}}{\bar{X}} \times 100 \quad \dots\dots\dots(8)$$

Where,

$\bar{X}$  = General mean of the character under study

$\sigma^2_g$  = Genotypic variance

##### 2.4.4. Phenotypic coefficient of variation (PCV %)

Phenotypic coefficient of variation was calculated using the following formula described in below.

$$PCV\% = \frac{\sqrt{\sigma^2_p}}{\bar{X}} \times 100 \quad \dots\dots\dots(9)$$

Where,

$\sigma^2_p$  = Phenotypic variance

$\bar{X}$  = General mean of the character under study

Classification of PCV and GCV were done following the method as suggested by Robinson et al. (1949).

<10% Low

10–20% Moderate

>20% High

##### 2.4.4. Heritability

The broad sense heritability ( $h^2_b$ ) was calculated for each traits by dividing genotypic variance and the phenotypic variance. The method followed was suggested by Johnson et al. (1955).

$$h^2_b (\%) = \frac{\sigma^2_g}{\sigma^2_p} \times 100 \quad \dots\dots\dots(10)$$

Where,

$h^2_b$  = Heritability (broad sense)

$\sigma^2_g$  = Genotypic variance

$\sigma^2_p$  = Phenotypic variance

Classification of heritability was done by following a method as suggested by Robinson et al. (1949).

<30% Low

30–60% Moderate

>60% High

##### 2.4.5. Genetic advance (GA)

It was measured the improvement rate in the mean of genotypes over the parental population. It could be calculated by using the methodology suggested by Johnson et al. (1955) at 5% selection intensity using the constant 'k' as 2.06.

$$GA = K \times h^2_b \times \sigma_p \quad \dots\dots\dots(11)$$

Where,

$h^2_b$  (bs) = Heritability in broad sense

$\sigma_p$  = Phenotypic standard deviation of the trait

K = Standard selection differential which is 2.06 at 5% selection intensity

#### 2.4.6. Genetic advance as % mean (GAM)

The genetic advance express as % of mean was calculated as per the formula suggested by Johnson et al. (1955).

$$GA (\% \text{ of mean}) = \frac{GA}{\bar{X}} \times 100 \quad \dots\dots\dots (12)$$

0–10%: Low

10–20%: Moderate

20% & above: High

#### 2.4.7. Test of normality

Skewness and kurtosis were calculated by the SPSS V20

#### 2.4.8. Correlation analysis

Correlation analysis was performed by using R software V4.3.1

### 3. Results and Discussion

Lab experiment was conducted at Department of Biochemistry, B.A.C.A, Anand Agricultural University, Anand, Gujarat, India. Out of total 28 genotypes, 14 highly resistant genotypes selected for biochemical analysis were 1 to 14 and remaining 14 genotypes, 15 to 28 were selected as susceptible. Both contrasting genotypes compared to the resistant parent AB 15-06 and susceptible parent GRB 5. Data on chlorophyll ( $\text{mg g}^{-1}$ ), moisture (%), total phenol ( $\text{mg g}^{-1}$ ), total soluble sugar ( $\text{mg g}^{-1}$ ), membrane injury (%) were analysed from young fresh leaves and results had been presented in (Table 1) with interpretation.

#### 3.1. Biochemical characterization of brinjal genotypes and their parents

##### 3.1.1. Chlorophyll content

Little leaf (phytoplasma) infection interferes with plant physiology in various manners and leading to the reduction of chlorophyll concentration. It blocks the regions that are used in chlorophyll production and as a result, the plant cannot produce pigment. At the same time, affected plant specimens often appear to be stunted and bear leaves with irregular shapes that decrease the planted area available for photosynthesis; therefore, there is a decrease in the demand for chlorophyll. However, there is also an aberration in vital nutrients which are essential in the synthesis and stability of chlorophyll because of derangement in nutrient provision. So for that reason, the chlorophyll level reduces substantially and photosynthesis process and overall health of the plant will be affected badly.

Resistant genotypes 1 to 14 contained chlorophyll in the ranged of 0.73 to 0.98  $\text{mg g}^{-1}$  while susceptible genotype 15 to 28 chlorophyll content with the ranged of 0.25 to 0.53  $\text{mg g}^{-1}$ , respectively. Resistant parent AB 15-06 chlorophyll contained 0.92  $\text{mg g}^{-1}$  which was higher as compared to susceptible parent GRB 5 recorded 0.40  $\text{mg g}^{-1}$ . Compare to the resistant

parent AB 15-06, genotype 12 (0.98  $\text{mg g}^{-1}$ ) and 13 (0.96  $\text{mg g}^{-1}$ ) recorded statistically higher value compare to susceptible parent GRB 5. Lowest chlorophyll content was observed in genotype 21 (0.25  $\text{mg g}^{-1}$ ). Meena et al. (2016) carried out biochemical characterization from health and chilli leaf curl infected leaves of chilli. Chlorophyll content measured in healthy leaves and infected leaves was 0.4932  $\text{mg g}^{-1}$  and 0.142  $\text{mg g}^{-1}$ , respectively. Singh et al. (2023) evaluated biochemical changes during Vegetable mite, *Tetranychus neocaledonicus* André affected brinjal leaves compare to healthy leaves. Total chlorophyll content estimated 3.86 $\pm$ 0.22  $\text{mg g}^{-1}$  in infected leaves compare to healthy leaves was 5.47 $\pm$ 0.32  $\text{mg g}^{-1}$ .

##### 3.1.2. Moisture

Brinjal little leaf disease causes a decrease in moisture content via a number of different processes. First, the disease causes plants to grow slowly and develop their leaves abnormally which lowers transpiration rates, the process by which water vapor is expelled from leaves. As there is less transpiration, there is less intake of water through the root system leading to a reduced amount of water throughout. However, infection with the phytoplasma affects the normal functions of the plant as a result of aberrations in metabolic activities within the plant. This disturbance may entail alterations to the absorption and retention characteristics of water, which would serve to lower the moisture content even more.

In present study, results revealed that resistant genotypes (1 to 14) contained moisture in the ranged of 88.63% to 97.43%, while susceptible genotypes (15 to 28) contained the range of moisture 76.77% to 83.43%. In resistant parent AB 15-06 moisture content 96.53% which was higher as compared to susceptible parent GRB 5 which had 84.93% moisture content. The lower moisture content 76.77% recorded in 17 and higher moisture content 97.43% was observed in genotype 6 which noted statistically higher moisture content over susceptible parent.

##### 3.1.3. Total phenol

Little leaf disease infection in brinjal, the plants own defense mechanisms lead to an increase of the total Phenol content. Thus, phenolic compounds have the function of antioxidants and antimicrobial agents, which are very important for defense against pathogens. They also function as chemical messengers in the body, synchronising the body's activities to the infection. Strengthening the cell wall and impeding pathogen spread are pivotal roles of phenolic compounds. The disruption of normal metabolic pathways leads to the accumulation of phenols, indicating a resource reallocation towards bolstering the plant's defense. The increase in total phenol content emphasizes the intricate relationship between plant and pathogen and represents the plant's coordinated attempt to ward off little leaf disease.

The ranged from resistant parent AB 15-06 contained total phenol 0.89  $\text{mg g}^{-1}$  which was lower as compared to susceptible



Table 1: Biochemical analysis of brinjal genotypes and their parents from leaves

Genotype	CHY (mg g <sup>-1</sup> )	MOI (%)	PHL (mg g <sup>-1</sup> )	TSS (mg g <sup>-1</sup> )	MI (%)	DI (%)
1	0.73	89.50	1.02	3.52	28.56	0.00
2	0.86	95.03	0.95	2.45	30.32	0.00
3	0.77	94.87	0.87	3.71	33.28	0.00
4	0.83	97.03	1.20	3.29	24.56	0.00
5	0.80	90.80	1.15	2.36	25.32	0.00
6	0.84	97.43	0.43	2.96	31.21	0.00
7	0.74	94.00	1.02	2.41	29.44	0.00
8	0.92	88.63	1.08	3.48	26.63	0.00
9	0.81	93.03	0.88	3.31	27.02	0.00
10	0.84	93.87	1.66	3.59	26.59	0.00
11	0.92	91.67	1.20	4.65	28.01	0.00
12	0.98	92.13	1.10	4.27	32.04	0.00
13	0.96	94.77	1.21	4.57	28.10	0.00
14	0.86	89.30	1.67	4.15	26.42	0.00
15	0.53	78.20	2.29	7.74	69.88	38.30
16	0.34	83.73	2.85	6.67	63.50	36.90
17	0.52	76.77	2.87	5.35	54.44	32.14
18	0.40	83.43	2.97	5.55	55.25	32.14
19	0.49	77.20	2.69	5.58	56.83	32.14
20	0.43	82.23	2.83	7.27	58.07	32.14
21	0.25	78.97	2.23	5.28	57.21	32.54
22	0.47	79.80	2.93	7.36	58.63	35.14
23	0.39	82.80	3.02	6.45	64.56	35.89
24	0.55	79.60	2.47	6.28	52.63	30.90
25	0.27	82.63	2.34	5.55	58.55	32.14
26	0.45	78.93	3.26	5.26	68.46	37.50
27	0.40	83.07	2.81	6.58	66.86	37.58
28	0.47	78.37	2.50	6.85	64.35	36.12
AB 15-06 (P <sub>1</sub> )	0.92	96.53	0.89	1.81	24.55	0.00
GRB 5 (P <sub>2</sub> )	0.40	84.93	3.16	7.68	69.91	34.50
SEm±	0.01	0.22	0.03	0.08	0.91	1.07
CD (p=0.05)	0.02	0.62	0.08	0.22	2.58	2.97
CV %	1.83	4.38	2.70	2.81	3.51	24.82

Note: CHY: Chlorophyll; MOI: Moisture; PHL: Phenol; TSS: Total soluble sugar; MI: Membrane injury; DI: Disease incidence

parent GRB 5 3.16 mg g<sup>-1</sup>. Minimum phenol content was observed in 0.43 mg g<sup>-1</sup> in genotype 6 and maximum phenol content 3.26 mg g<sup>-1</sup> was observed in genotype 26. In resistant genotypes (1 to 14) contained phenol in the ranged of 0.43 mg g<sup>-1</sup> to 1.67 mg g<sup>-1</sup>, while susceptible genotypes (15 to 28) contained the ranged of phenol 2.50 mg g<sup>-1</sup> to 3.26 mg g<sup>-1</sup>. Venkataravanappa et al. (2022) identified biochemical

changes during little leaf infection in brinjal. The total phenol content was increased in little leaf infected leaves 349.2 to 567.21 mg 100 g<sup>-1</sup> compare to the healthy leaves was 133.72 to 785.12 mg 100 g<sup>-1</sup>. Moderately resistant genotypes observed higher phenol content than susceptible variety. Singh et al. (2023) evaluated biochemical changes during Vegetable mite, *Tetranychus neocaledonicus* André affected brinjal





leaves compare to healthy leaves. Total phenol content was increase  $8.48 \pm 1.30 \text{ mg } 100 \text{ g}^{-1}$  in infected leaves compare to uninfected leaves was  $7.14 \pm 1.37 \text{ mg } 100 \text{ g}^{-1}$ . The results are in harmony with the findings Kumar et al. (2023) also carried out biochemical characterization of phenol ranges in between 1.50 to  $3.30 \text{ mg g}^{-1}$  in brinjal.

#### 3.1.4. Total soluble sugar

During little leaf infection plant in stress condition, thus activates the stress response that results to the production of soluble sugars. These sugars are osmotically active, they help keeping the cellular osmotic pressure and shield the cells from damage due to dehydration. Also, in stress conditions plants redistribute resources for the management of the stress stimulus. Phytoplasma infection, plant prefer to produce and stored soluble sugars as a sign of defense. They also act as important energy providers of different glycolytic pathways for both reaction to the pathogen and other cellular metabolic functions. Also, since normal metabolic processes are altered due to the infection hence carbohydrate metabolism soluble carbohydrates appear in higher concentrations as metabolic intermediates are by passed or accumulated due to the infection.

The resistant parent AB 15-06 contained total soluble sugar  $1.81 \text{ mg g}^{-1}$  which was lower as compared to GRB 5 susceptible parent  $7.68 \text{ mg g}^{-1}$ . In resistant genotypes (1 to 14) contained total soluble sugar in the ranged of  $2.36$  to  $4.57 \text{ mg g}^{-1}$ , while susceptible genotypes (15 to 28) contained the ranged of total soluble sugar between  $5.28$  to  $7.36 \text{ mg g}^{-1}$ . Minimum  $2.45 \text{ mg g}^{-1}$  and maximum  $7.74 \text{ mg g}^{-1}$  total soluble sugar content was observed in 2 and 15 genotypes of brinjal, respectively. Meena et al. (2016) carried out biochemical characterization from healthy and chilli leaf curl infected leaves of chilli. Total soluble sugar content measured in healthy leaves and infected leaves was  $3.60 \text{ mg g}^{-1}$  and  $3.90 \text{ mg g}^{-1}$ , respectively.

#### 3.1.5. Membrane injury

Membrane injury typically higher in susceptible parents compared to resistance parents when plants are exposed to pathogens. Susceptible parents usually possess weaker defense mechanisms. In contrast, resistance parents have stronger defense mechanisms against pathogens.

The higher membrane injury 69.91% was observed in susceptible parent GRB 5 and lower membrane injury 24.55% recorded in resistant parent. Lower membrane injury 24.56% was observed in resistant genotype 4 whereas higher membrane injury 69.88% was found in susceptible genotype 15. Results revealed that resistant genotype (1 to 14) contained membrane injury in the ranged of 24.56% to 33.28% while susceptible genotype (15 to 28) contained the ranged of 52.63% to 69.88%. Genotype 15 (69.88%) and 26 (68.46%) membrane injury were noted statistically higher over susceptible parent. The present results are in conformity with Gobu et al. (2017) carried out membrane injury in brinjal. Range varies between 24.33% to 66.66% in both healthy and

stressed related brinjal plants however results remained in contradiction with Mahammed et al. (2021) where membrane injury were recorded between 10.49% to 46.27% in total 50 genotypes of brinjal plant.

#### 3.2. Correlation study of disease Incidence with biochemical traits

The pearson's correlation (Figure 1) revealed a significant negative association between disease incidence with chlorophyll content ( $r=-0.93^{***}$ ), moisture ( $r=-0.91^{***}$ ) were as positively correlate with total soluble sugar ( $r=0.88^{***}$ ), membrane injury ( $r=0.93^{***}$ ) and phenol ( $r=0.94^{***}$ ). Chlorophyll content was significantly negative associated with disease incidence ( $r=-0.93^{***}$ ), phenol ( $r=-0.86$ ), membrane injury ( $-0.86^{***}$ ), total soluble sugar ( $r=-0.76^{***}$ ) were as moisture ( $r=0.83^{***}$ ) positively correlate with chlorophyll. Moisture content significant negative correlate with disease incidence ( $r=-0.91^{***}$ ), phenol, membrane injury and total soluble sugar ( $r=-0.87^{***}$ ,  $r=-0.83^{***}$  and  $r=-0.81^{***}$  respectively). Total soluble sugar positively correlate with disease incidence, phenol, membrane injury ( $r=0.88^{***}$ ,  $r=0.86^{***}$ ,  $r=0.83^{***}$ ). Membrane injury positively correlate with disease incidence and phenol ( $r=-0.93^{***}$  and  $r=0.91^{***}$ ).

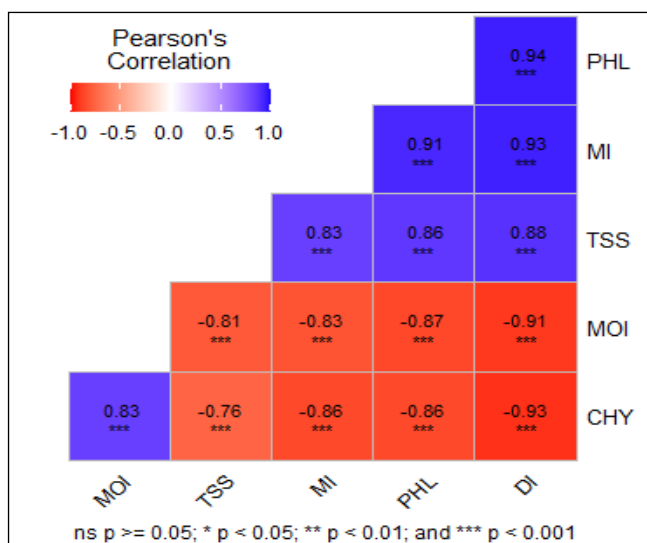


Figure 1: Correlation coefficient analysis of biochemical parameters with disease incidence of brinjal genotypes; Note: CHY: chlorophyll; MOI: moisture; PHL: phenol; TSS: total soluble sugar; MI: Membrane injury

Results indicated that characters like chlorophyll, membrane injury moisture were observed significant negative correlation with disease incidence while rest of biochemical parameters like total soluble solids and phenol content showed positive correlation.

##### 3.2.1. Test of normality

Discrete variation in the population is expressed by quantitative characters. For a given characteristic, skewness and kurtosis were computed to determine the frequency distribution of

brinjal genotypes and their genetic relationships.

The frequency distribution curve for the biochemical features of the brinjal genotypes displayed in Figure 2. Skewness and kurtosis also measured from brinjal genotypes showed in Table 2.

Table 2: Skewness and kurtosis for biochemical traits of brinjal genotypes

Sr. No.	Traits	Skewness	Kurtosis
1.	Chlorophyll	-0.079	-1.318
2.	Membrane injury	0.254	-1.540
3.	Moisture	0.026	-1.543
4.	TSS	0.030	-1.160
5.	Phenol	0.016	-1.626

Biochemical characters like membrane injury (0.254), moisture (0.026), total soluble sugar (0.030), phenol (0.016) showed positive skewness. This indicates that a greater number of genotypes than would be predicted from a normal distribution are below the mean whereas chlorophyll (-0.079) detected negative skewness. This indicated that more genotypes that would be predicted from a normal distribution are above the mean.

For kurtosis, all biochemical traits like chlorophyll (-1.318), membrane injury (-1.540), moisture (-1.543), total soluble sugar (-1.160), phenol (-1.626) detected negative kurtosis which means average level of complementary gene activity though to be platykurtic that indicated the presence of numerous small gene progressively greater effects controlled activity of the genes.

### 3.3. Variability analysis of biochemical traits from brinjal genotypes

The variability among various traits is assessed using the co-efficient of variation that is computed at the phenotypic and genotypic levels. Heritability provides an estimate of the relative amount of heritable portion of variation, while GCV and PCV indicated the existence of the moderate to high values were observed which potential for scope for improvement through selection (Deshmukh et al., 1986).

Heritability value in combination with high genetic advancement provides an accurate estimate of the amount of genetic advancement resulting from the selection of the finest individuals reported by Burton and Devane (1953) and Johnson et al., 1955. Variability analysis for biochemical traits of brinjal genotypes were mentioned in Table 3.

#### 3.3.1. Chlorophyll

Chlorophyll ranged in between 0.2 to 1.0 mg g<sup>-1</sup> was observed along with closely association of GCV (36.46%) with PCV (36.50%) which suggested less role of environment. The high estimates of heritability (99.82%) with high values % mean of genetic advance (75.04%) provided the additive genes effect

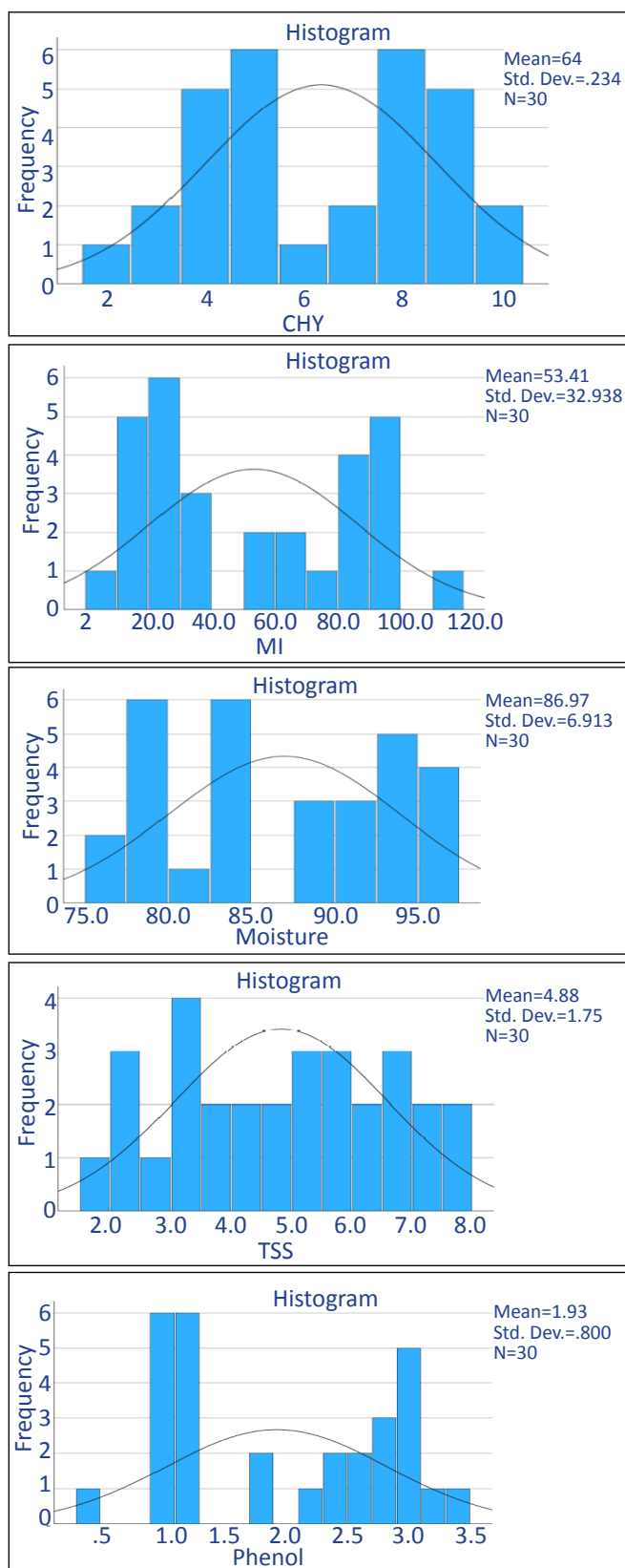


Figure 2: Frequency distribution of biochemical characters of brinjal genotypes; Note: CHY: Chlorophyll; MI: Membrane injury; TSS: Total soluble sugar



Table 3: Variability analysis for biochemical traits of brinjal genotypes

Sl. No.	Traits	Range	Mean	GCV (%)	PCV (%)	H <sup>2</sup> b (%)	GAM (%)
1.	chlorophyll (mg g <sup>-1</sup> )	0.2–1.0	0.64	36.46	36.5	99.82	75.04
2.	membrane injury (%)	9.7–98.33	53.41	61.52	61.96	98.59	125.84
3.	moisture (%)	76.8–97.4	8.7	7.55	8.73	74.75	13.44
4.	total soluble sugar (mg g <sup>-1</sup> )	1.8–7.7	4.87	35.89	36	99.39	73.7
5.	phenol (mg g <sup>-1</sup> )	0.4–3.3	1.92	46.76	46.83	99.67	96.15

and less environmental influence for the inheritance of the trait and selection may be effective. High GCV (20.91%) and PCV (20.94%) with high heritability (97.00%) and % mean of genetic advance (42.99%) were recorded by Das et al. (2018) in tomato.

### 3.3.2. Membrane injury

Membrane injury ranged between 9.7 to 115.5% was observed along with closely association of GCV (61.52%) and PCV (61.96%) which suggested the presence of good amount of variability among the genotypes for trait. Presence of high variability indicated less environmental influence hence selection can found to be rewarding. The high estimates of heritability (98.59%) with high values % mean of genetic advance (125.84%) indicated the influence of additive genes and low environmental influence and the selection will be rewarding for crop improvement for this trait.

### 3.3.3. Moisture

Moisture ranged 76.8 to 97.4% was observed along with closely association of GCV (7.55%) and PCV (8.73%) which suggested less variation among the genotypes for moisture content. The high estimates of heritability (74.75%) with moderate values % mean of genetic advance (13.44%) which indicated which is express preponderance non-additive gene effect in the inheritance of this trait which indicated fair chances for improvement in this trait for following selection.

### 3.3.4. Total soluble sugar

Total soluble sugar ranged in between 1.8 to 7.7 mg g<sup>-1</sup> was observed along with closely association of GCV (35.86%) with PCV (36.00%) were high, which indicated the existence of high variability for total soluble sugar. Least difference between GCV and PCV values indicated little contribution of the environment for the expression of the trait. The high estimates of heritability (99.39%) with high values % mean of genetic advance (73.70%) which suggested the influence of additive gene and selection would be effective for this trait. High GCV (24.85%) and PCV (24.67%) with high heritability (99.0%) and % mean of genetic advance (116.27%) were observed by Das et al. (2018) in tomato.

### 3.3.5. Phenol

Phenol content ranged in between 0.4 to 3.3 mg g<sup>-1</sup> had close association with GCV (46.76%) and PCV (46.83%) High values suggested the presence of good amount of variability

which may be utilized for selection in more effective breeding programme. The high estimates of heritability (99.67%) with high values % mean of genetic advance (96.15%) which suggested the importance of additive gene effect and direct selection for improvement of this trait in segregating generation of these population would be effective.

## 4. Conclusion

Resistant parent AB 15-06 was recorded chlorophyll, moisture content (from brinjal leaves) which was higher as compared to susceptible parent GRB 5. Correlation analysis for phenol, total soluble sugar and membrane injury showed significant positive correlation with the disease incidence. Normality distribution analysis for membrane injury, moisture, total soluble sugar, phenol were governed by the complementary gene action whereas chlorophyll governed by the duplicate gene action. GCV and PCV% found highest in membrane injury and phenol content from brinjal leaves.

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