Morphological and biochemical screening of upland cotton (Gossypium hirsutum L.) hybrids for jassid (Amrasca devastans Dist.) resistance

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Abstract

The current experiment was carried out in the field in a Randomized Block Design (RBD) for parents viz., MCU5, MCU7, CO14, and CO17, which were utilized as lines (high yield) and KC2, KC3, NDLH1755 and NDLH1938, which were used as testers throughout the summer season of 2021. From the mean values among all parents calculated, KC 2 had lowest jassid population of mean 4.13 numbers per leaf, highest amount of phenol (3.72 mg/g), low amount of reducing sugars (1.22 mg/g) and high chlorophyll index (58.73). Among other hybrids the hybrid MCU 5 × KC 3 outperformed for highest phenol content (3.72 mg/g), highest chlorophyll index (58.73) with lowest Jassid count (4.13 numbers/plant). Hence from the correlation results, number of jassids per plant would be lower for high phenolic content, low reducing sugar, lower soluble proteins, higher chlorophyll index and higher trichome density. Hence breeding could be made for resistance to jassids by insight into this biochemical analysis and trichome density of the cultivar which would help reduce use of insecticides and pest load from the fields thereby enhancing the yield of the population.

Keywords: Jassid screening, biochemical, phenol, reducing sugar, trichome length, trichome density, soluble proteins

Introduction

Cotton (Gossypium hirsutum L.), a valuable crop is a major industrial commodity across the world. It is the second-oldest and most important commercial crop. India accounts for 33% (10.7 million hectares) of world cotton acreage and 22% (5.4 million tonnes) of global cotton production. The unsystematic application of chemical pesticides to control major pests on cotton led to development of pesticide resistance in both targeted and non-targeted pests, disruption of the natural balance of pests and their natural enemies, resurgence of minor pests, pollution of the crop ecosystem, health and economic risks and development of sucking pest resistance (Pawar and Kadam 1995) [17]. Furthermore, conventional pesticides do not provide the intended control of sucking pests and leafhoppers and whitely outbreaks on cotton are common (Ahmad et al. 2002) [13].

Eco-friendly pest management is required in an integrated approach to ensure a sustainable production. Despite the availability of effective insecticides and the drawbacks of chemical insecticide use, cultivating resistant cotton cultivars is the cheapest and most harmless strategy to managing sucking pest infestations in an integrated pest management programme. Breeders can use the resistant cultivar in their breeding programme to produce resistant genotypes that are environmentally sustainable, socially stable, and economically viable. As a result, resistant cotton cultivars play an important role in a long-term agricultural system. Expertise is required to collect data on specific biochemical features and their relationship to sucking pest infestation (Rizwan et al. 2021) [19]. The number of sucking pests varies from cultivar to cultivar due to physical characteristics of cotton plants, particularly on leaves. Biochemical components such as protein, nitrogen, phosphorus, potassium, phenol, and sugar are among the most well-known characteristics associated with resistance. Potential biotic stressors are expressed by insects to their host plants. Plants that are attacked by insects respond by changing the composition and characteristics of their cell walls, as well as the content of their nutrients (secondary metabolite biosynthesis) (Hopkins and Huner, 2004) [9]. Modifications in plant protein profiles and oxidative enzymes have been shown to be among a plant’s earliest responses to insect herbivores (Rafie et al. 1996; Chaman et al. 2001 and Ni et al. 2001) [8, 16].
As a result, the current research will focus on the many biochemical (phytochemical) compounds found in plants that are important for conferring resistance/susceptibility to sucking pest in chosen cotton cultivars.

Materials and Methods
The current experiment was carried out in the field in a Randomized Block Design (RBD) for parents, MCU5, MCU7, CO14, and CO17, which were utilized as lines (high yield) and KC2, KC3, NDLH1755 and NDLH1938, which were used as testers throughout the summer season of 2021 (testers parent for jassid resistance and lines selected had good quality for lint). Using Dock's method, each of the four lines was crossed with four testers individually utilizing manual emasculation and pollination (Doak, 1934) [6]. In 2021, 16 hybrid F1 seeds were raised, along with their 8 parents and a standard check (MCU 5). The genotypes were then raised in Department of cotton, Tamil Nadu Agricultural University, Coimbatore (2020-2021).

Total soluble proteins
Total soluble protein in cotton leaves was estimated using Lowrey et al. 1951 [13]. Using a known concentration of bovine serum albumin, a standard curve was created. Protein content in both types of samples was estimated using a standard calibrated curve with known bovine protein values.

Total reducing sugars: With a few adjustments, the total reducing sugars were computed using the Miller et al. (1972) approach. The total reducing sugars of the hybrids and parents were estimated using a standard graph of glucose created by known glucose concentrations (100-500 g).

Table 1: Show the symptoms

| Grades | Symptoms |
|--------|----------|
| 1. | Leaves free from crinkling or with no yellowing, bronzing and drying |
| 2. | Few leaves on lower portions of the plant curling, crinkling and slight yellowing |
| 3. | Crinkling and curling all over, yellowing, bronzing and browning in the middle and lower portion, plant growth hampered; and |
| 4. | Extreme curling, yellowing, bronzing and browning, drying of leaves and defoliation, stunted growth |

Where G represented the number of the grade of ICCC and P the number of plants under each entry. Grouping of injury index to categories of resistance was as follows.

| Grade index | Category |
|-------------|----------|
| 0.0–1.0     | Resistant |
| 1.1–2.0     | Moderately resistant |
| 2.1–3.0     | Susceptible |
| 3.1–4.0     | Highly susceptible |

Trichome density and Trichome length analysis
The MI-DC1300, MI-DC3000, and MI-DC5000 are compound microscope digital cameras that provide crisp images with excellent contrast and colour. The photos for the leaf samples that were newly harvested at 50 DAS and sliced to the size of one centimetre square were presented on the slides using the Scope Photo software. Then still shots were taken, and the analysis performed.

Chlorophyll index
A KONICA MINOLTA SPAD – 502 machine was used to determine it. The SPAD metre is a useful tool for measuring chlorophyll index fast and non-destructively. The measurement was saved to memory automatically. The SPAD meter results were proportional to the quantity of chlorophyll in the leaf, and they were calculated by measuring the leaf absorbance in the red and near-infrared bands (Uddling et al., 2007) [24].

Result and Discussion
Mean performance of parents and hybrids
The parents MCU 5, MCU 7, CO 14 and CO 17 were used as lines with good fibre quality traits; KC 2 KC3, NDLH 1755 and NDLH 1938 were used as testers with resistance to Jassid. MCU 5 had highest yield of 117.69 g/plant among all parents. Among the lines, CO 17 had maximum mean jassid population per leaf (9.69) with lowest amount of phenol (1.25 mg/g), highest amount of reducing sugar (3.87 mg/g) and lowest chlorophyll index (22.43). KC 2 had lowest jassid population of mean 4.13 numbers per leaf, highest amount of phenol (3.72 mg/g), low amount of reducing sugars (1.22 mg/g) and high chlorophyll index (58.73). MCU 7 among the lines had maximum injury grade of 2.40 reported as moderately resistant and maximum trichome length of 1.70 mm per square centimetre. The tester NDLH 1938 had lowest injury grade of 0.36 with the lowest trichome length of 0.49 mm per square centimetre. The tester KC3 had highest trichome length of 1.73 mm per square centimetre and lowest
trichome density of 115.15 numbers per square centimeter. The hybrid MCU 5 X KC 2 noted for highest phenol content (3.47 mg/g) with resistant injury grade of less than one and highest seed cotton yield per plant (155.32 g/plant) indicating trichome density indirectly paves way for jassid resistance thereby increasing seed cotton yield of the plant. The hybrid MCU 7 X NDLH 1755 had lowest mean of seed cotton yield per plant (82.78 g/plant) among the hybrids with susceptible grade. Shinde et al. (2014) [22] also reported high phenolics provide resistance to plants from Jassids. Higher phenolic content along with higher trichome density in cotton plant would result in a highly resistant plant was also suggested by Rohini et al. (2011) [20]. As phenol content in cotton plants acts as the feeding deterrent the population of jassid feeding that respective plant would be lower. This result was in accordance with Manivannan et al. (2021) [19]. Similar result of higher protein favouring higher number of jassid population was also reported by him. Higher amount of reducing sugar would result in susceptible genotypes for jassid. Hence breeding could be made for resistance to jassids by insight into this biochemical analysis and trichome density of the cultivar which would help reduce use of insecticides and pest load from the fields thereby enhancing the yield of the population.

**Correlation**

Number of jassids per plants showed highly positive significance with amount of reducing sugars (0.914) but negatively correlated with trichome density (-0.835), amount of phenols (-0.857) present in the plant and chlorophyll index (-0.871). Ahmad et al. (2005), Ashfaq et al. (2010) [3], Rustamani et al. (2014) [21], Gonde et al. (2015) [8], Kanher et al. (2016) [12], Sankeshwar et al. (2016) [22] and Amin et al. (2017) [2] also reported negative correlation of number of jassids with trichome density. Trichome length was found to have non significant positive correlation with number of jassids which is not in agreement with Batti et al. (2015) [4]. Injury grade showed high negative correlation with amount of phenol (-0.652) and chlorophyll index of the plant (-0.490). The phenol content present in the plant was highly negatively correlated with amount of reducing sugars (-0.849), amount of soluble proteins present (-0.432). Amount of reducing sugar is highly negatively correlated with the chlorophyll index (-0.914). Seed cotton yield of the plant had non-significant correlation with number of jassids per plant (-0.021) and pest index score (-0.023).

Hence from the above results, number of jassids per plant would be lower for high phenolic content, low reducing sugar, lower soluble proteins, higher chlorophyll index and higher trichome density. The phenol content present in the plant was highly significantly correlated with amount of reducing sugars (0.914), amount of soluble proteins present (-0.432). Amount of reducing sugar is highly negatively correlated with the chlorophyll index (-0.914).

### Table 2: Mean performance of parents for Jassid population biochemical and trichome studies along with the yield parameter

| Crosses | Mean Jassid nymph/plan t (Nos.) | Jassid injury grade | Trichome length (mm/sq.cm) | Trichome density (nos./sq.cm) | Total phenol (mg/g) | Total reducing sugar (mg/g) | Total soluble protein (mg/g) | Chlorophyll index | Seed cotton yield per plant (g/plant) |
|---------|---------------------------------|---------------------|-----------------------------|-------------------------------|---------------------|-----------------------------|-----------------------------|-------------------|-------------------------------------|
| Lines   |                                 |                     |                             |                               |                     |                             |                             |                   |                                     |
| MUC5    | 9.28 ab                         | 1.31 cde            | 1.58 defg                   | 115.151                       | 1.38 e              | 3.62 a                      | 36.42 bcd                   | 28.10 hij          | 102.83 ghf                           |
| MUC7    | 9.49 a                          | 2.40 b              | 1.70 cdef                   | 154.89 ijk                    | 1.32 e              | 3.64 a                      | 32.79 ef                    | 25.78 ij           | 90.53 ghij                           |
| CO14    | 9.49 a                          | 1.75 bc             | 1.44 efghi                  | 148.91 jkl                    | 1.32 e              | 3.79 a                      | 35.51 cd                    | 23.74 hjk          | 89.05 hjk                            |
| CO17    | 9.69 a                          | 1.29 cde            | 1.23 hijk                   | 145.60 jkl                    | 1.25 e              | 3.87 a                      | 30.15 gh                     | 22.43 k            | 83.22 ij                             |
| Testers |                                 |                     |                             |                               |                     |                             |                             |                   |                                     |
| KC2     | 4.13 i                          | 0.73 defg           | 0.92 kl                     | 273.96 abc                    | 3.72 a              | 1.22 g                      | 32.71 ef                    | 58.73 a            | 79.24 j                              |
| KC3     | 4.14 i                          | 0.69 defg           | 1.73 bcde                   | 262.31 bcd                    | 3.65 a              | 1.30 g                      | 27.46 i                     | 57.47 a            | 94.67 fghi                            |
| NDLH 1755 | 4.14 i                         | 0.74 defg           | 1.26 ghj                    | 290.66 ab                     | 3.47 a              | 1.42 g                      | 37.32 abc                   | 56.20 a            | 88.51 hj                             |
| NDLH 1938 | 4.54 hi                        | 0.36 g              | 0.49 m                      | 238.15 cde                    | 3.45 a              | 1.43 g                      | 32.12 fg                    | 54.66 ab           | 90.17 ghj                            |
| Mean    | 6.86                            | 1.16                | 1.30                        | 203.70                        | 2.45                | 2.54                        | 33.06                       | 40.89              | 103.42                               |
| Max     | 9.69                            | 2.40                | 1.73                        | 290.66                        | 3.72                | 3.87                        | 37.32                       | 58.73              | 117.69                               |
| Min     | 4.13                            | 0.36                | 0.49                        | 115.15                        | 1.25                | 1.22                        | 27.46                       | 22.43              | 89.82                                |

* Same alphabetical letters indicate denotes they were non-significant.
Table 3: Mean performance of 16 hybrids for Jassid population, biochemical and trichome studies along with the yield parameter

| Crosses          | Mean Jassid nymph/ plant (Nos.) | Jassid injury grade | Trichome length (mm/sq.cm) | Trichome density (nos/sq.cm) | Total phenol (mg/g) | Total reducing sugar (mg/g) | Total soluble protein (mg/g) | Chlorophyll index | Seed cotton yield per plant (g/plant) |
|------------------|---------------------------------|---------------------|-----------------------------|-------------------------------|---------------------|-----------------------------|-----------------------------|-------------------|-------------------------------------|
| MCU5 X KC 3      | 4.65 h                          | 1.63 e              | 1.38 fghi                   | 304.80 a                       | 3.42 a              | 1.50 g                      | 30.45 fgh                   | 52.53 abc         | 106.28 fg                            |
| MCU5 X NDLH 1755 | 5.15 h                          | 1.16 cdef           | 0.54 m                      | 262.10 bcd                    | 3.39 a              | 1.76 fg                     | 27.34 i                      | 48.61 bc          | 144.56 ab                            |
| MCU5 X KC 2      | 7.85 cdef                       | 3.85 a              | 1.17 hijkl                   | 271.42 abc                    | 1.68 e              | 2.30 de                     | 31.19 fg                     | 44.48 def         | 124.10 de                            |
| MCU5 X NDLH 1938 | 7.62 defg                       | 3.52 a              | 2.16 a                      | 200.94 fgh                    | 1.50 e              | 2.31 de                     | 36.89 bcd                    | 39.88 hj          | 106.35 fg                            |
| MCU7 X KC 2      | 7.35 efg                        | 3.37 a              | 1.28 ghij                    | 183.38 ghij                   | 1.59 e              | 3.32 ab                     | 35.47 cd                     | 30.10 hj          | 124.96 cde                           |
| MCU7 X KC 3      | 7.20 efgh                       | 3.34 a              | 1.01 jkl                     | 153.17 ijk                    | 1.73 de             | 2.90 bc                     | 39.56 a                      | 32.72 gh           | 128.60 bcd                           |
| MCU7 X NDLH 1755 | 7.15 efgh                       | 3.17 a              | 1.96 abc                     | 184.88 ghij                   | 1.77 cde             | 2.82 bcd                    | 36.62 bcd                    | 33.22 gh           | 82.78 ij                             |
| MCU7 X NDLH 1938 | 6.88 fg                         | 3.12 a              | 1.92 abc                     | 208.36 efgh                   | 2.32 bc              | 2.66 cde                    | 38.63 ab                     | 39.13 efg         | 136.90 bcd                           |
| CO14 X KC 2      | 6.78 fg                         | 1.77 bc             | 1.50 defgh                   | 227.06 defgh                  | 2.32 bc              | 2.66 cde                    | 37.51 abc                    | 38.59 fg          | 110.51 ef                            |
| CO14 X NDLH 1755 | 6.55 g                          | 1.35 cde            | 2.03 ab                      | 239.96 efgh                   | 2.64 b              | 2.64 cde                    | 28.66 hi                     | 40.57 ef          | 152.90 a                             |
| CO14 X NDLH 1938 | 5.17 hij                        | 0.27 g              | 1.23 hijk                    | 204.55 efgh                   | 3.38 a              | 1.77 fg                      | 32.80 ef                     | 46.49 cde         | 155.32 a                             |
| CO17 X KC 2      | 8.12 bede                       | 0.61 efg            | 2.19 a                       | 129.30 kl                     | 2.69 a              | 2.60 cde                    | 31.48 fg                     | 40.79 ef          | 134.28 bcd                            |
| CO17 X NDLH 1938 | 8.56 abedc                      | 0.40 fg             | 1.13 ijk                     | 169.02 hjj                    | 1.76 a              | 3.32 de                     | 32.30 efg                    | 41.52 def         | 97.14 fgh                             |
| CO17 X NDLH 1755 | 5.37 h                          | 1.90 bc             | 0.87 i                       | 275.35 abc                    | 3.37 a              | 2.14 ef                     | 27.46 i                      | 45.70 cdef        | 92.44 ghij                            |
| Mean             | 6.88                            | 1.95                | 1.51                         | 212.66                        | 2.43                | 2.49                         | 33.66                         | 40.47             | 123.11                                |
| Max              | 8.90                            | 3.85                | 2.19                         | 304.80                        | 3.42                | 3.49                         | 39.56                         | 52.53             | 155.32                                |
| Min              | 4.65                            | 0.27                | 0.54                         | 129.30                        | 1.38                | 1.50                         | 27.34                         | 29.88             | 82.78                                 |

Table 4: Simple correlation of Jassid population, biochemical and trichome analysis along with the yield parameter

|      | NJP  | IG    | TL    | TD    | P     | RS    | SP    | CI    | SCY   |
|------|------|-------|-------|-------|-------|-------|-------|-------|-------|
| NJP  | 1    |       |       |       |       |       |       |       |       |
| IG   | 0.273| 1     |       |       |       |       |       |       |       |
| TL   | 0.375| 0.191 | 1     |       |       |       |       |       |       |
| TD   | -0.835** | -0.069 | -0.341 | 1     |       |       |       |       |       |
| P    | -0.857** | -0.652** | -0.37 | -0.799** | -0.849** | 1     |       |       |       |
| RS   | .914** | 0.324 | 0.353 | -0.799** | -0.849** | 1     |       |       |       |
| SP   | 0.269 | 0.366 | 0.298 | -0.374 | -0.432 | 0.342 | 1     |       |       |
| CI   | -0.871** | -0.490** | -0.353 | -0.792** | -0.921** | -0.797 | 1     |       |       |
| SCY  | -0.021 | -0.023 | 0.181 | -0.013 | 0.043 | 0.014 | 0.018 | 0.03  | 1     |

**Correlation is significant at the 0.01 level (2-tailed).
*Correlation is significant at the 0.05 level (2-tailed).

NJP - Mean number of Jassids per leaf, S - Amount of reducing sugars (mg/g), TL - Trichome length (mm), IG - Pest Injury Grade, SP - Total soluble proteins (mg/g), TD - Trichome Density (nos./cm²), P - Total amount of phenol (mg/g), CI - Chlorophyll index, SCY - Seed cotton yield per plant
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