Research Article

Varietal resistance of Bt cotton against sucking complex under field conditions at upper Sindh-Pakistan

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Abstract
The research work was carried out during Kharif season, 2018 to investigate the varietal resistance of six Bt cotton varieties i.e., FH-Lalazar, SS-32, Bt-CRIS-508, CRIS-510, FH-142 and CRIS-533 against sucking pests, jassid, whitefly, thrips and mealybug under field conditions. The maximum resistivity against jassids was observed in SS-32 (1.98) per leaf followed by Bt-CRIS-508 (2.01), CRIS-533 (2.10), CRIS-510 (2.11), FH-142 (4.02) and FH-Lalazar (4.06). Against whitefly SS-32 gave maximum resistance (5.99) per leaf followed by CRIS-533 (6.00), CRIS-510 (8.55), Bt-CRIS-508 (8.93), FH-142 (9.12) and FH-Lalazar (11.04). Against thrips SS-32 variety showed maximum resistance (8.57) per leaf followed by CRIS-510 (8.81), CRIS-533 (8.87), Bt-CRIS-508 (9.50), FH-142 (10.27) and FH-Lalazar (15.31), while against mealybug Bt-CRIS-508 found resistive (0.42) followed by CRIS-510 (0.43), FH-142 (1.49), SS-32 (1.88), CRIS-533 (1.98) and FH-Lalazar (2.02). The overall results showed that cotton variety SS-32 gave better resistance against overall sucking pests (4.60) per leaf followed by CRIS-533 (4.74), and CRIS-510 (4.97), while varieties Bt-CRIS-508 and FH-142 showed moderate resistance against sucking pests (5.21) and (6.23), respectively however; variety FH-Lalazar had not shown enough resistance (8.11) against all the four mentioned sucking pests, so cotton variety FH-Lalazar was considered as most susceptible Bt variety and SS-32 variety was found to be most resistive variety against all the four sucking pests except mealybug. So it is suggested that the variety SS-32 should be cultivated as it is less attacked by sucking pests.

Keywords: Bt cotton; Sucking pests; Vitro conditions

Introduction
Cotton (Gossypium hirsutum L.) is a very important crop containing fiber and maintaining cash. In 2017 the growing area of cotton crop in Pakistan was 2.489 million hectares and its production was 10.671 million bales [1]. It is commercial, historical, industrial and cultural crop, its silver fiber attracts world’s interest. In Pakistan it earns a substantial foreign exchange of 68% by exporting raw cotton and 7.1% in agriculture value addition and about 1.6% to GDP [2]. It is attacked by various sucking and chewing insect pests but major sucking complex among them are jassid, Amrasca biguttula biguttula (Ishida) whitefly, Bemisia tabaci (Gennadius), thrips, Thrips tabaci (Lindeman), mealybug, Phenacoccus solenopsis (Tinsley) and aphid, Aphis gossypii (Glover) which are cause of the most damage to the cotton crop. Cotton jassid is the notorious sucking insect pest of cotton [3, 4]. This pest mainly found on lower side of plant leaves, sucks sap from lower side portion of the leaves and injects the poisonous substance into the plant tissues. So, plant leaves become wrinkled and this is the symptom of jassid infestation [5]. Cotton whitefly also lives underside of cotton leaves, sucks the cell sap,
causes 50% reduction of the production of the bolls [6], also it is vector of famous disease of cotton crop namely cotton leaf curl virus (CLCuV) [7]. There are about 600 host plants of whitefly, which is menacing to agriculture-based countries [8]. Thrips are tiny insect pests which live lower side of cotton leaves and gave harm to plant by sucking sap of the cell, due to that, the leaves of the cotton plant become cup-shaped and of silvery appearance [3]. Mealybug is a small soft-bodied pest, which found mostly on the stem and leaves of the plant and sucks the cell sap [9]. Its adult female is covered over by waxy powder due to this; its common name is mealybug [10]. Its hosts are different like harvests, vegetables, decorative plants and weeds [11], it gave 14% loss to cotton yield, recently it become serious pest of cotton [12]. These sucking pests are playing major role in reducing cotton production throughout the Pakistan [13, 14]. Due to massive usage of pesticides, these pests got high resistance and the environment becomes polluted along with different health risks [15, 16].

From 1996 world-renowned that Bt cotton is the definitive solution to overcome the resitvity problems against chewing pests. An enormous resistivity has been adopted by chewing pests such as Helicoverpa armigera, Pectinophora gossypiella, Earias vittella and Earias insulana against Bt cotton under field and laboratory conditions [17]. Therefore, without using insecticides, varietal resistance tactic is of huge importance [18, 19, 20] for developing the pest management strategy, it is main requirement to understand the effect of different morphological characters of plants for the host selection behavior. In Pakistan, the breeders have evolved a number of varieties by focusing their concentration to enhance the yield capacity. Plant pests and natural enemies can be positively or negatively affected by many plant characters [21, 22]. In Pakistan the preceding efforts were taken to investigate the tactics to increase host plant resistance to insect pests in cotton; like those of [15, 20, 23, 24], etc, but still a lot remains yet to be completed to reach some specific outcomes. Keeping in view; the importance of resistant varieties the present studies were conducted to screen out 6 cotton genotypes. The natural resistances against these sucking complexes were also determined. Thus, the main object was to observe the population fluctuation for most resistant and susceptible Bt cotton varieties against sucking insect pests.

**Materials and methods**

The experimental trial was conducted during Kharif season, 2018 to investigate the resistivity of Bt cotton varieties against sucking complex i.e., jassid, whitefly, thrips and mealybug under field conditions. For this purpose the seeds of six different Bt cotton varieties i.e., T1= FH-Lalazar, T2=SS-32, T3= Bt-CRIS-508, T4=CRIS-510, T5= FH-142 and T6= CRIS-533 were sown on ridges in North to South direction in a Randomized Complete Block Design (RCBD) having a treatment size of 100 sq feet, which were replicated 4 times and kept without using of insecticides. The space of 2 feet was maintained between treatments and replications by separating each plot, while 18” distance was maintained between the ridge to ridge and 9” to 12” distance between plant to plant. Thinning, weeding and other agronomical practices were manually done. Before sowing the cotton crop, the pre-weedicide Stomp (FMC Pvt. Ltd. Pakistan) was applied for controlling the weeds.

When the sucking insect pests had made their initial appearance sporadically, the crop was checked for the presence of the sucking insect pests on a weekly basis. Twenty plants of each cotton variety were chosen, randomly per treatment/plot for recording pest population. Three leaves, one each from the bottom, middle and top portions of cotton plants were kept under observation for insect pest population. The information was obtained by the average counts of the pest per leaf. The varietal resistance was observed during the season of the most insect pest activity.

**Statistical analysis**

The information on the individual population of each sucking complex species was displayed through a different examination of the mean
values. The methods were isolated by observing the Least Significant Difference (LSD) test at (P<0.05) through Statistics (8.1) computer software, student package, USA. The relationship framework between the overall and individual population dynamics of the sucking complex on various cotton cultivated varieties was analyzed.

Results
As the infestation of the pest is to be reciprocal of the insect pest susceptibility, so with an increase in pest population per leaf, the relative resistance of the variety was considered to decrease as reported by Aslam et al. [25].

Resistance of Bt cotton varieties to jassid
The population of jassid started from the month of July when plant bore 5-6 leaves after one month of cultivation of the cotton crop under field conditions which remained active up to the end of September or harvesting of the crop. All the six cotton varieties were cultivated on the same date and at the same location which was infested by the sucking complex vigorously. From which cotton jassid pest was observed with the overall maximum mean population (4.06) per leaf on FH-Lalazar under field conditions (Table 1). This variety had maximum number of jassids followed by FH-142 (4.02), CRIS-510 (2.11), CRIS-533 (2.10) and Bt-CRIS-508 (2.01) per leaf throughout the cotton crop season but the least number of jassids were found on SS-32 (1.98) per leaf that found comparatively resistant when compared to the other tested varieties.

Further, the ANOVA showed a significant difference in two groups (DF= 11, 5; F= 75.93; P= 0.001) against jassid population, one of them included FH-Lalazar and FH-142, while the second group included CRIS-533, CRIS-510, Bt-CRIS-508, and SS-32, respectively.

Resistance of Bt cotton varieties to whitefly
The population of whitefly on all the six Bt varieties of cotton was observed and it was found that SS-32 variety was relatively more resistant to whiteflies among the tested varieties, as it is shown in (Table 2) per leaf least number of whiteflies (5.99) followed by CRIS-533 (6.00) whiteflies. The FH-Lalazar was found to be the most vulnerable as it showed maximum whiteflies per leaf (11.04), followed by FH-142, Bt-CRIS-508 and CRIS-510 having (9.12), (8.93) and (8.55) whiteflies respectively. Overall results showed that FH-Lalazar was significantly different (DF= 11, 5; F= 13.24; P= 0.001) from all other varieties containing the maximum number of whiteflies while Bt-CRIS-508, CRIS-510, FH-142 were statistically similar and CRIS-533, SS-32 were also similar to each other for whiteflies per leaf.

Resistance of Bt cotton varieties to thrips
The population of thrips was also observed on all the six tested varieties of cotton, the results revealed that SS-32 showed maximum resistance to thrips comparatively to other tested varieties (Table 3). It showed least number of thrips (8.57) per leaf followed by CRIS-510 (8.81), CRIS-533 (8.87). Similarly; the maximum number of thrips was found on FH-Lalazar (15.31) per leaf followed by FH-142 (10.27) and Bt-CRIS-508 (9.50). All the varieties were significantly different (DF= 11, 5; F= 76.97; P= 0.001) from one another except CRIS-510 and CRIS-533 which were non-significant to each other.

Resistance of Bt cotton varieties to mealybug
The population of mealybug appeared late in the 2nd week of August on all the six tested varieties of the cotton crop as shown in (Table 4), among these varieties Bt-CRIS-508 and CRIS-510 showed maximum resistance to mealybug (0.42), (0.43) respectively as compared to other tested varieties. While all the other four varieties FH-142, SS-32, CRIS-533, and FH-Lalazar were containing almost similar mean population of mealybug (1.49), (1.88), (1.98) and (2.02) respectively. Among all these tested varieties FH-Lalazar was found less resistant to mealybug as compared with other varieties. The ANOVA showed the significant difference (DF= 11, 5; F= 5.97; P= 0.001) among all tested Bt varieties.
### Table 1. Overall mean population of cotton jassid under field conditions during 2018

| Name of Variety\Weeks | Months | July | August | September | Mean± S.E |
|-----------------------|--------|------|--------|-----------|----------|
| FH Lalazar            |        |      |        |           |          |
|                       |        |      |        |           |          |
| SS-32                 |        |      |        |           |          |
| Bt-CRIS-508           |        |      |        |           |          |
| CRIS-510              |        |      |        |           |          |
| FH-142                |        |      |        |           |          |
| CRIS-533              |        |      |        |           |          |

Means followed by common letters in the respective category are not significantly different from each other by LSD at $\alpha = 0.05$.

### Table 2. Overall mean population of cotton whitefly under field conditions during 2018

| Name of Variety\Weeks | Months | July | August | September | Mean± S.E |
|-----------------------|--------|------|--------|-----------|----------|
| FH Lalazar            |        |      |        |           |          |
|                       |        |      |        |           |          |
| SS-32                 |        |      |        |           |          |
| Bt-CRIS-508           |        |      |        |           |          |
| CRIS-510              |        |      |        |           |          |
| FH-142                |        |      |        |           |          |
| CRIS-533              |        |      |        |           |          |

Means followed by common letters in the respective category are not significantly different from each other by LSD at $\alpha = 0.05$. 

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### Table 3. Overall mean population of cotton thrips under field conditions during 2018

| Name of Variety | Weeks | July | August | September | Mean± S.E |
|-----------------|-------|------|--------|-----------|-----------|
|                 | 1st   | 2nd  | 3rd    | 4th       | 5th       | 6th       | 7th       | 8th       | 9th       | 10th      | 11th      | 12th      |
| FH Lalazar      | 5.50  | 6.40 | 9.33   | 12.33     | 15.93     | 17.40     | 21.06     | 24.00     | 19.00     | 17.46     | 15.40     | 15.31±1.68 |
| SS-32           | 2.76  | 2.86 | 3.40   | 5.26      | 7.33      | 9.06      | 12.86     | 13.33     | 14.53     | 12.00     | 10.80     | 8.60      | 8.57±1.23  |
| Bt-CRIS-508     | 1.30  | 2.26 | 3.46   | 6.13      | 8.06      | 9.26      | 11.40     | 14.26     | 17.13     | 16.00     | 13.53     | 11.20     | 9.50±1.55bc |
| CRIS-510        | 0.54  | 1.20 | 2.26   | 4.26      | 7.53      | 10.93     | 11.46     | 13.93     | 16.66     | 15.00     | 12.00     | 10.00     | 8.81±1.61cd |
| FH-142          | 2.24  | 3.20 | 5.26   | 7.20      | 10.26     | 11.80     | 13.80     | 15.93     | 16.60     | 14.33     | 12.26     | 10.39     | 10.27±1.39b |
| CRIS-533        | 0.46  | 1.26 | 2.40   | 4.13      | 7.73      | 10.80     | 13.06     | 15.26     | 16.00     | 15.33     | 11.00     | 9.00      | 8.87±1.64cd |

Means followed by common letters in the respective category are not significantly different from each other by LSD at α = 0.05.

### Table 4. Overall mean population of cotton mealybug under field conditions during 2018

| Name of Variety | Weeks | July | August | September | Mean± S.E |
|-----------------|-------|------|--------|-----------|-----------|
|                 | 1st   | 2nd  | 3rd    | 4th       | 5th       | 6th       | 7th       | 8th       | 9th       | 10th      | 11th      | 12th      |
| FH Lalazar      | 0.00  | 0.00 | 0.00   | 0.00      | 0.00      | 0.54      | 0.93      | 1.33      | 2.80      | 4.00      | 6.00      | 8.64      | 2.02±0.82a  |
| SS-32           | 0.00  | 0.00 | 0.00   | 0.00      | 0.00      | 0.66      | 0.96      | 1.46      | 2.40      | 3.33      | 5.93      | 7.80      | 1.88±0.75a  |
| Bt-CRIS-508     | 0.00  | 0.00 | 0.00   | 0.00      | 0.00      | 0.00      | 0.26      | 0.53      | 0.80      | 0.93      | 1.13      | 1.40      | 0.42±0.15b  |
| CRIS-510        | 0.00  | 0.00 | 0.00   | 0.00      | 0.00      | 0.00      | 0.33      | 0.54      | 0.76      | 0.96      | 1.20      | 1.38      | 0.43±0.15b  |
| FH-142          | 0.00  | 0.00 | 0.00   | 0.00      | 0.00      | 0.33      | 0.86      | 1.17      | 2.73      | 3.05      | 4.15      | 5.60      | 1.49±0.56a  |
| CRIS-533        | 0.00  | 0.00 | 0.00   | 0.00      | 0.00      | 0.60      | 1.03      | 1.73      | 3.20      | 4.33      | 5.66      | 7.17      | 1.98±0.73a  |

Means followed by common letters in the respective category are not significantly different from each other by LSD at α = 0.05.
Resistance of Bt cotton varieties to overall sucking pests

The resistance level of tested Bt cotton varieties against different sucking complex is shown in (Fig. 1). It was observed that cotton variety SS-32 showed better resistance to overall sucking pests (4.60) mean population per leaf followed by CRIS-533 (4.74) and CRIS-510 (4.97), while varieties Bt-CRIS-508 and FH-142 showed moderate resistance to all sucking pests (5.21) and (6.23) mean population per leaf respectively; however, variety FH-Lalazar had not shown enough resistance (8.11) against all the four mentioned sucking pests, so it was considered as most susceptible among the tested transgenic varieties. Further, the ANOVA showed that against sucking pests all the varieties were significantly different (DF= 11, 5; F= 65.74; P= 0.001) from one to another except SS-32 and CRIS-533 which were non-significant to each other. Means followed by common letters in the respective category are not significantly different from each other by LSD at α = 0.05.

Weekly population fluctuation of sucking complex

The results of the population fluctuation of all the four sucking complex on various transgenic cotton varieties are shown in (Fig. 2) which illustrates that infestation of the jassid remained above economic threshold level (ETL) almost throughout experimental period except 1st week of data collection and its peak population was found during 3rd week of August which is (3.94) per leaf. While the attack of whitefly remained below ETL in starting; during the first three weeks of July then gradually increased and crossed the ETL during the month of August and its population even more increased in September reached at peak (13.46) per leaf during 2nd week of September. Likewise mean population of thrips on various Bt cotton varieties remained below ETL in July then its population crossed the ETL
and reached at peak (17.49) per leaf in the last week of August then its population declined slowly but remained above ETL. As for as population fluctuation of mealybug is concerned on various Bt cotton varieties; it did not appear till the 1st week of August on the all tested varieties of cotton crop, after appearance gradually its population increased day by day from the 2nd week of August till the end of September. Further, the ANOVA showed the overall significant difference (DF= 5, 11; F= 128.68; P= 0.001) against sucking pests among all the twelve weeks (throughout the season) from one to another except 9\textsuperscript{th} and 10\textsuperscript{th} week, which were non-significant to each other.

![Figure 2. Weekly infestation ratio of sucking complex on Bt cotton varieties throughout the season](image_url)

**Discussion**

In current research trial, six Bt cotton varieties were cultivated which were infested by the sucking complex vigorously, from which cotton jassid pest was observed with the overall per leaf maximum mean population (4.06) on FH-Lalazar under field conditions followed by FH-142 (4.02), CRIS-510 (2.11), CRIS-533 (2.10) and Bt-CRIS-508 (2.01) per leaf throughout the cotton crop season but the least number of jassids were found on SS-32 (1.98) per leaf that was found comparatively resistant to sucking pests when compared to other tested varieties. Our results are in similarity with Saleem et al. [28] who also found maximum jassid population on FH-142 variety of Bt cotton. Sucking complex was found dangerous for the cotton crop, controlled by many methods among them varietal resistance of cotton is a major achievement in agriculture research. According to [29] the impact of cotton varieties on insect pests may be different and resistivity of cultivated varieties to insect pests could play a major role to obtained insect pests reduction biologically and cultivation varieties resistant to insect pests could play effective role to combat insect pests biologically. While population of whitefly on six Bt cotton varieties were observed among these varieties SS-32 variety was found relatively resistant to whiteflies, as it showed per leaf minimum number of whiteflies (5.99)
followed by CRIS-533 (6.00). Whereas; FH-Lalazar was found to be the most vulnerable to whiteflies as it showed maximum number of whiteflies per leaf (11.04), followed by FH-142 (9.12), Bt-CRIS-508 (8.93) and CRIS-510 (8.55). Our results are in resemblance with [26] who found maximum whiteflies on FH-Lalazar, and also in analogy with him on FH-142 because he reported less number of whiteflies on FH-142, while in similarity with [27] who also reported that FH-Lalazar is most susceptible to whiteflies and who also found infestation of whiteflies on FH-142 on second number as compared to other genotypes of cotton. The results about thrips population on tested Bt cotton varieties exposed that maximum resistance against thrips was observed by cotton variety SS-32, which showed per leaf minimum number of thrips (8.57) followed by CRIS-510 (8.81), CRIS-533 (8.87). While per leaf the maximum number of thrips were observed on FH-Lalazar (15.31) followed by FH-142 (10.27) and Bt-CRIS-508 (9.50). These current results are in similarity with [27] who found maximum infestation of thrips on FH-Lalazar, while in analogy with [26] who found minimum number of thrips on FH-Lalazar and FH-142 and also in analogy with [28] who found minimum number of thrips on FH-142 variety of Bt cotton. The results about mealybug infestation on six tested varieties of Bt cotton revealed that two varieties Bt-CRIS-508 and CRIS-510 showed maximum resistance against mealybug (0.42), (0.43) respectively; followed by FH-142, SS-32, CRIS-533 and FH-Lalazar showed mean population of mealybug (1.49), (1.88), (1.98) and (2.02) respectively. Among all these tested varieties FH-Lalazar was found less resistive to mealybug as compared with other varieties. Our results are in analogy with [30] who described the non-resistivity of Bt cotton against sucking complex i.e., jassids, whiteflies, thrips, aphids and mealybugs, who also said that pesticides were used to control them.

The overall resistance level of six Bt cotton varieties against different sucking pests was observed, in which cotton variety SS-32 showed better resistance to overall sucking pests (4.60) mean population per leaf followed by CRIS-533 (4.74) and CRIS-510 (4.97), while varieties Bt-CRIS-508 and FH-142 showed moderate resistance to all sucking pests (5.21) and (6.23) mean population per leaf respectively; however, variety FH-Lalazar had not shown enough resistance (8.11) against all the four mentioned sucking pests, so it was considered as most susceptible Bt variety among the tested transgenic varieties. Our results are supported by [31] who reported that there was no significant difference between Bt and non-Bt cotton varieties in having population of jassid, whitefly and thrips, further also supported by [32, 33] who described that Bt cotton has no effect on population of non-target sucking complex so there is need of regular integrated pest management practices.

The results of population fluctuation of all the four sucking complex on six transgenic cotton varieties illustrated that infestation of the jassid remained above economic threshold level (ETL) almost throughout experimental period except for 1st week of data collection and its peak population was found during 3rd week of August which is (3.94) per leaf. Our findings are in similarity with [28, 31, 34] who also found peak point of jassid population in the month of August but with little difference between 1st and 3rd week. While the attack of whitefly remained below ETL in starting; during the first three weeks of July then gradually increased and crossed the ETL during the August and its population even more increased in September reached at peak (13.46) per leaf during 2nd week of September. Our findings are in difference with [4, 28, 31] who described that the whitefly population reached its peak in month of August. Likewise mean population of thrips on various Bt cotton varieties remained below ETL in July then its population crossed the ETL and reached at peak (17.49) per leaf in the last week of August then its population declined slowly but remained above ETL. Our findings are in similarity with [4, 31] they also described that thrips population reached a peak in month of
August, further support in similar types of results by [35-37]. Our results are different from [28] who found that thrips population was started to gradually build up from last week of June and reached on peak in 1st week of August and then started declined. As for as population fluctuation of mealybug on various Bt cotton varieties is concerned; it did not appear till the 1st week of August on the all tested varieties of cotton crop then its population increased day by day from the 2nd week of August till the end of September. Our results are in similarity with [38] who described that sucking pest mealybug has emerged on cotton crop after 2 months of cultivation and its population was found till the harvest of the crop.

**Conclusion**

Sucking complex, jassid, whitefly, thrips, and mealybug are more dangerous to the cotton crop. Their population was checked on six Bt cotton varieties under field conditions among them cotton variety FH-Lalazar was found to be more susceptible and cotton variety SS-32 gave the good resistivity against sucking pests. So the variety SS-32 should be cultivated for better production of cotton.

**Authors’ contributions**

Conceived and designed the experiments: HA Sahito, Performed the experiments: ZH Shah, Analyzed the data: ZH Shah, Contributed materials/analysis/tools: HA Sahito, Wrote the paper: ZH Shah.

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