J. Cotton Res. Dev. **31** (2) 283-289 (July, 2017) **ISSN No. 0972-8619**

Screening of *Bt* cotton hybrids against sucking pests (whitefly and leafhopper) attack and cotton leaf curl virus disease

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ABSTRACT : Sucking pests (whitefly and leafhopper) and cotton leaf curl virus disease (CLCuD) pose serious threat to cotton crop in south western region of Punjab. Whitefly (*Bemisia tabaci*) act as a major vector for transmission of cotton leaf curl virus. The trial was conducted to screen the *Bt* cotton hybrids along with check against the sucking pest and cotton leaf curl virus disease (CLCuD) for two consecutive years. During 2012, the population of whitefly ranged from 4.33 (in RCH 650 BGII) to 29.00 (in BCH 1545 BGII) adults/ 3 leaves and the population of leafhopper ranged from 2.00 (in DPC 3083BGII, NCCH 316BGII and NCS 855BGII) to 4.33 (66 SS 33BGII) adults/ 3 leaves. The minimum per cent disease index was 13.9 (RCH 650BGII) whereas maximum was 80.6 (66 SS 33BGII). In 2013, the population of whitefly was higher as compared to 2012 and ranged from 14.66 (DPC 3083BGII) to 49.00 (45 SS 33 BGII)/ 3 leaves whereas that of leafhopper ranged from 3.99 (45 SS 33BGII) to 11.33 (in 54 SS 33BGII)/ 3 leaves. The minimum per cent disease index was 28.6 (DPC 3081BGII), higher than that in 2012 whereas maximum per cent disease index was 69.0 (in VBCH 1545BGII). The study showed that probably there is a correlation between population of whitefly and disease severity (R^2 for 2012 = 0.59 and for 2013 = 0.76) as with the increase in pest population, the genotypes became more susceptible to the disease though one hybrid (DPC 3081BGII) remained moderately resistant. The experiment conducted revealed that the hybrids which were found to be resistant to cotton leaf curl virus disease in the year 2012 lost their resistance in 2013 became susceptible to this disease except hybrid DPC 3081BGII and one check (NCS 855BGII) which remained tolerant in the 2nd year. The seed cotton yield was also recorded highest in DPC 3081BGII.

Key words : *Bt* cotton hybrids, cotton leaf curl virus disease (CLCuD), leafhopper, non *Bt* cotton hybrids, seed cotton yield, whitefly

Gossypium species, is one of the important *kharif* crops popularly known as "White Gold". Cotton is an extensively studied crop and is the chief source of natural fibre worldwide (Riaz *et al.,* 2013). The crop is of great economic importance as it plays a vital role in agricultural and industrial development and enables to earn foreign exchange through export of its raw materials as well as finished products (Tuteja, 2014). The initiation of symptoms of cotton leaf curl virus disease (CLCuD) are characterized by small vein thickenings (SVT) type symptom on young upper leaves of plants followed by upward/ downward leaf curling and leaf thickening. Infected plants later develop leafy enations on the underside of leaf and become stunted leading to severe reduction in the yield and the disease is transmitted by its vector, whitefly, *Bemisia tabaci* (Monga, 2014). These *Bt* cotton hybrids are adversely affected by the cotton leaf curl virus disease and sucking pests *i.e.* whitefly, leafhopper etc which is emerging as a major constraint in cotton production. The disease and pest attack not only reduce the yield but also affect the quality of the fibre. Cotton leaf curl virus disease (CLCuD) is amongst the major

factor for the low productivity and adversely affects the yield and quality characters of upland cotton. (Singh *et al.,* 2013).The fibre properties play a vital role in assessing the quality of cotton(Siwach *et al.,* 2016). To manage the cotton leaf curl virus disease and sucking pest (whitefly, Jassid) attack, the key measures that should be taken are strict enforcement to ban CLCuD *Bt* cotton Hybrids and susceptible varieties in North India, ensuring cultivation of tolerant/ resistant genotypes (Kranthi, 2014), identification of resistant genotypes, chemical control of whitefly and rouging of infected plants and weeds (Monga, 2014). A large number of *Bt* cotton hybrids are developed by different private seed companies are being cultivated by the farmers of our country. It is the need of the hour for the screening of the *Bt* cotton hybrids against disease/pests attack before recommending to the farmers. With this objective, some of the *Bt* cotton hybrids were screened out against the disease/pest attack and yield under irrigated conditions of south western region of Punjab.

MATERIALS AND METHODS

Experimental site : The experiment was conducted at Regional Research Station, Bathinda, Punjab for two consecutive years *i.e.* 2012 and 2013. In this study, 15 *Bt* cotton hybrids evaluated in PAU for population of sucking pests(whitefly and leaf hopper) and severity of cotton leaf curl disease . The crop was sown at a site having 74° 18' E longitude, 30° 58' N latitude and an altitude of 211 m at 67.5 × 75cm spacing. Each hybrid was replicated thrice in a randomized block design. At different phases of the crop, all the recommended fertilizers, irrigation and cultural practices were given for full potential assessment with respect to yield.

Data recording : The data on various aspects *i.e.*, population of whitefly and leafhopper, cotton leaf curl disease severity and seed cotton yield was recorded from 15 randomly selected plants in all the three replications. The standard protocol was used to assess the incidence of CLCuD, the detail of which is given in Table 1.

Table 1. Level of infestation and reaction of CLCuV

Disease Severity (Grade)	Disease Index (%)	Disease Reaction
1	0	Immune/Disease free
2	0.1 – 10	Highly resistant
2	10.1 – 20	Resistant
3	20.1 – 30	Moderately resistant
4	30.1– 40	Moderately susceptible
5	40.1 – 50	Susceptible
6	More than 50	Highly susceptible

Note: The CLCuD infestation data were recorded at 15 days interval throughout the season.

The data for sucking insect pest *viz.,* whitefly and leafhopper was recorded with reference to population. Five plants were randomly selected in each replication; hence the sample size became 15 plants which found suitable for analysis of the data. The data of the sucking insect pest was recorded after every 15 days till the maturity of the crop. The data was recorded from 3 leaves of a single plant and was taken before 10 am. Regarding yield attributes, the data of different parameters were recorded but only seed cotton yield (q/ha) is presented in this study.

Statistical analysis : The data recorded was subjected to analysis with CPCS1 software developed by Department of Mathematics and Statistics, Punjab Agricultural University, Ludhiana. The mean and critical difference was

calculated for finding the significance of the recorded data. The correlation between whitefly population and CLCuD was calculated with the Microsoft excel. The equation for the correlation coefficient is as under ;

$$Correl(X,Y) = \frac{\sum (x-\bar{x})(y-\bar{y})}{\sqrt{\sum (x-\bar{x})^2 \sum (y-\bar{y})^2}}$$

Where $\bar{x}$ and $\bar{y}$ are the samples mean average (array1) and average (array2).

The disease severity of CLCV was recorded by using different grades and these grades were converted into the percentage disease index.

$$\text{Percentage Disease Index} = \frac{\text{Sum of numerical ratings}}{\text{Total number of leaves observed} \times \text{maximum disease grade}} \times 100$$

RESULTS AND DISCUSSION

Whitefly population : The data for population of whitefly was taken on 15 randomly selected plants as no. of adults/ 3 leaves for two consecutive years 2012 and 2013 and is shown in Table 2. During 2012, whitefly population was highest in hybrid VBCH 1545BGII (29 adults/ 3 leaves) followed by KSCH 218BGII (26 adults/3 leaves). Lowest population was recorded in hybrid RCH 650BGII (4.33adults/ 3 leaves) which was even lower than both the checks *i.e.* LHH 144(6.33 adults/ 3 leaves) and NCS 855BGII (9/ 3 leaves). The whitefly population for all other hybrids was found between 6.33 to 24.33 adults/ 3 leaves. During 2013, the population of whitefly was found higher as compared to that in 2012. It reached to 49.00 adults/ 3 leaves in hybrid 45 SS 33 BGII and was highest of all the fifteen hybrids followed by 47 adults/ 3 leaves in hybrid Ankur 3028 BGII. The population was lowest in DPC 3081 BGII (14.66 adults/ 3 leaves) except non *Bt* check LHH 144 in which it was 9.66 adults/ 3 leaves. The whitefly population for all other hybrids ranged between 21.66 42.66 adults/ 3 leaves. It was observed that population of whitefly was lower in hybrid DPC 3081 BGII during both the years. Studies on whitefly population on infestation of 64 varieties have also been done by Parveen *et al.,* (2010).

CLCuD reaction and PDI : The data on cotton leaf curl virus disease severity was taken and percent disease index (PDI) was calculated and the subsequent disease reaction thereof was recorded for all the 15 hybrids along with *Bt* and non *Bt* check and is presented in Table 2. During 2012, most of the hybrids showed highly resistant reaction with PDI ranging between 10 20 except 3 hybrids 45 SS 33 BGII, KSCH 209 BGIl and 66 SS 33 BGII which were found highly susceptible to CLCuD with per cent disease index of 80.6, 58.3 and 50, respectively. The hybrid VBCH 1548 BGII was found tolerant with disease index of 25 per cent. Least per cent disease index was found in 3 hybrids namely RCH 650 BGII, DPC 3083 BGII, 51 SS 33 BGII and one check NCS 855 BGII. However, the non *Bt* check LHH 144 was found disease free, therefore was highly resistant. During 2013, per cent disease index was found higher for all the hybrids as compared to 2012. Nine hybrids showed highly susceptible reaction with PDI above 50.0 per cent *viz.,* VBCH 1548 BGII (69%) , MRC 7017 BGII (66.7%) , 45 SS 33 BGII (57.1%), KSCH 218 BGII (57.1%) , 54 SS 33 BGII(57.1%), 66 SS 33 BGII(57.1%), Ankur 3028 BGII(52.4%), Ankur 3244 BGII(50%) and NCCH 0316 BGII(50%). Five hybrids were moderately susceptible to CLCuD *i.e.* DPC 3083 BGII, KSCH 209 BGII and VBCH

Table 2. Average population of whitefly, leafhopper and yield during 2012 and 2013

Sr. N.	Entry	Whitefly/3 leaves			Leafhopper/3 leaves			Seed cotton yield (q/ha)		
		2012	2013	**Mean**	2012	2013	**Mean**	2012	2013	**Mean**
1	RCH 650 BG II	4.33	31.33	**17.83**	2.33	4.0	**3.16**	40.08	33.95	**37.01**
2	Ankur 3244 BGII	6.33	29.66	**17.99**	3.0	6.33	**4.66**	34.24	21.79	**28.01**
3	DPC 3081 BGII	8.33	14.66	**11.49**	2.99	6.0	**4.49**	43.71	31.41	**37.56**
4	DPC 3083 BGII	9.0	32.66	**20.83**	2.0	7.98	**4.99**	44.06	35.70	**39.88**
5	Ankur 3028 BG II	14.0	47.0	**30.5**	2.33	4.66	**3.49**	37.32	31.27	**34.29**
6	MRC 7017 BG II	7.66	42.66	**25.16**	2.99	8.0	**5.49**	39.5	28.04	**33.77**
7	NCCH 0316 BGII	11.0	28.33	**19.66**	2.0	4.33	**3.16**	32.61	28.54	**30.57**
8	45 SS33 BGII	21.0	49.0	**35**	2.33	3.99	**3.16**	18.81	15.53	**17.17**
9	KSCH 209 BGII	13.33	32.66	**22.99**	2.33	6.0	**4.16**	31.10	25.35	**28.22**
10	51SS33 BGII	14.0	34.0	**24**	3.66	7.33	**5.49**	20.23	21.69	**20.96**
11	KSCH 218BGII	26.0	39.66	**32.83**	4.0	9.66	**6.83**	26.42	25.69	**26.05**
12	54 SS33 BGII	24.33	37.33	**30.83**	3.33	11.33	**7.32**	25.49	16.16	**20.82**
13	VBCH 1548 BG II	18.66	30.0	**24.33**	2.66	8.0	**5.33**	36.04	23.46	**29.75**
14	VBCH 1545 BGII	29.0	29.66	**29.33**	3.0	4.33	**3.66**	39.80	24.86	**32.33**
15	66 SS33 BGII	21.66	22.33	**21.99**	4.33	5.66	**4.99**	12.30	23.01	**17.65**
16	NCS 855 BG II (Check)	9.0	18.66	**13.83**	2.0	4.33	**3.16**	41.20	30.48	**35.84**
17	LHH 144(Non *Bt*) (Check)	6.33	9.66	**7.99**	2.33	2.66	**2.49**	26.56	28.36	**27.46**
	CD (p=0.05)	**3.95**	**5.46**	**3.88**	**0.37**	**0.2**	**0.24**	**4.81**	**2.9**	**3.59**

1548 BGII with PDI =45.2, RCH 650 BGII and 51 SS 33 BGII with PDI = 40.5. Only one hybrid *i. e.* DPC 3081 BGII and one check NCS 855 BGII were found tolerant with PDI of 28.6. Non *Bt* check LHH 144 showed resistant reaction to the disease with PDI = 2.4.

From the above data, it is evident that the cultivars which showed moderately resistant reaction to the disease in 2012 became susceptible or highly susceptible in 2013. Only one hybrid *i.e.* DPC 3081 BGII which was resistant during 2012 do not lost resistance completely and became tolerant to the disease in 2013 which may be due to presence of certain genes in this cultivar the investigation for which is further needed to be done. Breakdown in resistance of different varieties released by state agricultural Universities has also been well studied by Monga (2014).

Relationship between whitefly population and CLCuD : The graph shown in the Fig.1 signifies that there is a positive correlation between whitefly population and cotton leaf curl virus disease for both the years (r^2 for 2012 =0.585, r^2 for 2013= 0.757). It is also concluded that with the increase in population of whitefly in 2013, cotton leaf curl virus disease severity is raised resulting in poor seed cotton yield. Disease severity also increases within the year with the increase in pest population. A single adult of whitefly is able to transmit leaf curl virus but the efficiency of transmission increases with the increase in population of its vector *Bemisia tabaci i.e.* number of adults more than 10/plant. In the present study also, it was observed that with the increase in number of whitefly, severity of cotton leaf curl increases as depicted by graph in Fig.1.

Leafhopper population : The data for leafhopper population was taken as adults

observed/ 3 leaves on 15 randomly selected plants for each hybrid for the year 2012 and 2013 (Table 4). In 2012, lowest leafhopper population was 2.00 adults/ 3 leaves observed in the hybrid DPC 3081 BGII , NCCH 0316 BGII and *Bt* check NCS 855 BGII. In four hybrids *viz.,* RCH 650 BGII, Ankur 3028 BGII, 45 SS 33 BGII and KSCH 209 BGII and one check LHH 144(non *Bt*) leafhopper population was 2.33 adults/ 3 leaves little higher than the lowest. Leafhopper population for other hybrids ranged between 2.99 4.33 adults/ 3 leaves. During 2013, leafhopper population was higher as compared to the previous year. It was lowest for 45 SS 33 BGII (3.99 adults/ 3 leaves) except check LHH 144 with a population of 2.66 adults/ 3 leaves and highest for 54 SS 33 BGII (11.33 adults/ 3 leaves). Leafhopper population for all other hybrids ranged between 4 to 9.66 adults/ 3 leaves.

From the above results, it is, therefore, concluded that there exists some relationship among the population of sucking pests, cotton leaf curl virus disease and seed cotton yield. It was observed that with the increase in leafhopper population in 2013, there is a decline in seed cotton yield. Therefore, it is concluded that leafhopper attack indirectly reduces yield and act as a limiting factor for cotton production. Reduction in yield with increase in jassid population is also well documented by Shivanna *et al.,* (2009).

Seed cotton yield : The data for seed cotton yield (SCY) was also taken for all the fifteen hybrids tested along with *Bt* and non *Bt* checks and is presented in Table 2. During 2012, three hybrids viz., DPC 3083 BGII, DPC 3081 BGII and RCH 650 BGII and one check NCS 855 BGII were found to have high yield potential and seed cotton yield was above 40q/ha and was 44.06,

Table 3. Per cent disease index and reaction of cotton leaf curl disease (CLCuD) on different cotton genotypes

Sr. No.	Entry	Cotton leaf curl virus disease			
		2012		2013	
		Per cent disease index	Reaction	Per cent disease index	Reaction
1	RCH 650 BG II	13.9	R	40.5	S
2	Ankur 3244 BGII	19.4	R	50.0	S
3	DPC 3081 BGII	16.7	R	28.6	MR
4	DPC 3083 BGII	13.9	R	45.2	S
5	Ankur 3028 BG II	19.4	R	52.4	HS
6	MRC 7017 BG II	16.7	R	66.7	HS
7	NCCH 0316 BGII	19.4	R	50.0	S
8	45 SS33 BGII	58.3	HS	57.1	HS
9	KSCH 209 BGII	50.0	HS	45.2	S
10	51SS33 BGII	13.9	R	40.5	S
11	KSCH 218BGII	19.4	R	57.1	HS
12	54 SS33 BGII	16.7	R	57.1	HS
13	VBCH 1548 BG II	16.7	R	45.2	S
14	VBCH 1545 BGII	25.0	MR	69.0	HS
15	66 SS33 BGII	80.6	HS	57.1	HS
16	NCS 855 BG II (Check)	13.9	R	28.6	MR
17	LHH 144(Non *Bt*) (Check)	0.0	HR	2.4	HR
	CD (p=0.05)	2.05		4.0	

43.71, 40.08 and 41.20q/ha, respectively. The seed cotton yield was lowest in hybrid 66 SS 33 BGII with SCY = 12.30 q/ha. The seed cotton yield for all other hybrids ranged between 18.81 39.80q/ha. In the year 2013, it was observed that seed cotton yield reduced subsequently for all the hybrids except 51 SS 33 BGII with SCY= 21.69q/ha and LHH 144 (check) with SCY = 28.36q/ha in which there was observed slight increase in seed cotton yield although SCY remained highest for DPC 3083 BGII (35.70q/ha) and lowest for hybrid 45 SS 33 BGII with SCY = 15.53q/ha. Seed cotton yield of DPC 3081 BGII, RCH 650 BGII, Ankur 3028 BGII and check NCS 855 BGII was 31.41,33.95,31.27 and 30.48 q/ha and was comparable to the highest (SCY = 35.70q/ha). Seed cotton yield for the rest of the hybrids remained between 16.16 to 28.54q/ha. The overall SCY for the two years was also highest in DPC 3083 BGII (SCY = 39.88 q/ha) and lowest for 45 SS 33 BGII. Seed cotton yield of different cotton cultivars also showed a decline in the year 2013. Reduction in yield was also observed (Monga, 2014 and Singh *et al.,* 2013).

In the present investigations, it was concluded that the hybrids which were resistant during 2012, resistance broken in 2013 and became susceptible or highly susceptible except hybrid DPC 3081 BGII in which population of whitefly and leafhopper was lower resulting in resistant and tolerant reaction to CLCuD in 2012 and 2013. The breakdown of resistance may be due to emergence of new strains of cotton leaf curl virus for which new source of resistance is needed to be incorporated. There is a need to develop cultivars which are resistant to sucking pest especially whitefly which is a limiting factor in cotton production by inciting cotton leaf curl viral disease. Probably some gene resistance may be needed to be investigated. Which may be present in diploid cultivars which are less attacked by sucking pests? These genes may possibly be transferred genetically to newly develop tetraploid cultivars for improving their disease and pest resistance. Some efforts has been made by workers (Nazeer *et. al.*, 2014) who explored the possibility of transferring genes for resistance to CLCuD from *Gossypium arboreum* (2n=26) cv 15 Mollisoni into *G. hirsutum* (2n=52) cv CRSM 38 through conventional breeding. Therefore, there is a great scope for developing resistance in agronomically suitable cotton cultivars in the near future. This information can further be implicated for finding out reasons for breakdown of resistance. Further work can be done in the field of biotechnology for

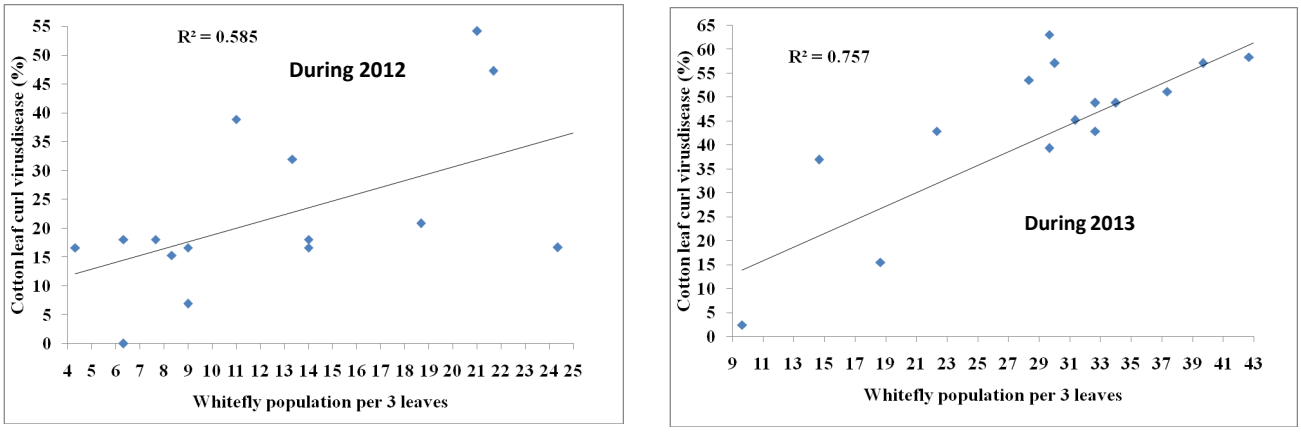

Fig. 1. Correlation (R^2) whitefly population and Cotton Leaf Curl Virus Disease

developing sucking pest and CLCuD resistant cotton cultivars. A little effort in this field can do wonders for the cotton growers.

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Received for publication : December 12, 2016
Accepted for publication : April 21, 2017