Chapter 14
Cotton Leaf Curl Virus Disease Status in *Bt* Cotton Hybrids in Punjab, India

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Abstract Cotton, popularly known as the "White Gold", is an important *Kharif* crop of the South Western Region of Punjab (India). *Bt* cotton refers to transgenic cotton, have an endotoxin protein inducing gene from soil bacterium *Bacillus thuringiensis*.

Insect-pests and diseases are the major biotic constraints in the production and productivity of the cotton crop in *Bt* and Non- *Bt* cotton varieties/hybrids. Among the diseases, viral disease i.e. Cotton leaf curl virus disease (CLCuD) is predominating in the cotton belt of Punjab in Northern India. Maximum areas of the cotton belt of the Punjab are under the *Bt* cotton hybrids in which an incidence of CLCuD are noticed although the disease severity varied in *Bt* and Non-*Bt* cotton varieties/ hybrids but the *Desi* cotton are found to be immune.

Variation in the disease severity might be due to prevalence of new virulent strains or combination of strains causing CLCuD in the changing environmental conditions. As per an information reported regarding strains from the literature i.e. Cotton leaf curl Burewala virus (CLCuBuV) which came into occurrence over a time, became a serious threat to the cotton cultivars not only in Punjab but throughout the Northern India. The attack of Cotton leaf curl virus disease (CLCuD) leads to frequent resistance breakdown among the *Bt* cotton hybrids and varieties. The 100% plants can be infected with the CLCuD having, with varying disease severity grade. Although, the plants infected in initial stages by CLCuD are more drastically affected the yield as compared to the plants infected at later stages. However, the CLCuD can be managed by adopting the integrated approaches to manage the vector (whitefly) of the CLCuD in cotton.

Keywords Cotton leaf curl virus disease (ClCuD) · *Bt* cotton hybrids/varieties · Resistance breakdown · Virulent strains · Time of sowing

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14.1 Introduction

When India became independent in 1947, the cotton scenario was hardly satisfying with production of 2.3 million bales as against mill consumption of 4.4 million bales of cotton, 40% of the most productive area going over to Pakistan and 95% of the mills remaining in India, were responsible for this gloomy scenario. Since the area, which went over to Pakistan was growing long and medium staple cotton, India suffered in quality front also. Extension efforts of the Governments of cotton growing states, with emphasis on extension of area and use of improved package of practices including use of adequate quantities of fertilizers, had salutary effect on both area and production of cotton. In less than a decade, the area and production stood at 8 million hectares and 4.2 million bales, respectively. During the next decade the production rose further to 5.3 million bales. There was no looking back thereafter and production continued to grow with area remaining static around 7.5 million hectares. It is at this point of time (1967) that the project widely known as AICRP was launched by ICAR(Indian Council of Agricultural Research) which gave great fillip to cotton research and its multidiscipline and Multilocation approach of improvement revolutionized in an enhancement in the seed cotton yield of cotton in a short span of time. The production and productivity of 5.3 million bales and 114 kg lint/ha in 1967 rose to 12 million bales and 280 kg ling/ha in 1991–92 respectively. Improved varieties and advent of hybrids, improved production and plant protection technologies, as well as extension efforts in disseminating these technologies have contributed to the progress in production and productivity. Qualitative improvement of cotton was also achieved due to development of varieties and hybrids of long and extra long staple (Paroda & Basu, 1992). Although, there is substantial increase in cotton production but an increase in yield is not as high as that for other crops.One of the important reason is due to the existence of the major biotic constraint in cotton crop i.e. Cotton leaf curl virus disease (CLCuD) transmitted by the whitefly. There were an number of reports in the literature regarding an incidence of CLCuD in all the cotton varieties and hybrids. None of the *Bt* cotton hybrids is noticed till date in Northern India which is immune to CLCuD. An epidemics of CLCuD in cotton crop was alsl noticed in the history. In this Book chapter, glimpse on the status of CLCuD in *Bt* cotton hybrids in Punjab in India will be noticed.

14.2 Hybrid Cultivars of Cotton

India is not only the first, but the only country who grow hybrid cotton on a large scale since seventies. It was at the Cotton Research Station, Surat that the first versatile hybrid H 4 (Hybrid 4) of intra hirsutum of cross (G-67 Am. Nectanless) was evolved. Similarly, hybrid Varalaxmi released in 1972 in India was the first interspecific (*hirsutum x barbadense*) cotton hybrid of the world.

The presence of high exploitable level of heterosis for seed cotton yield and its components, and the release of H4 and Varalaxmi hybrids for large scale cultivation in Central and South Cotton led to initiate systematic efforts for the development and testing of hybrids in North India. The main aim was to develop good quality and yield oriented hybrids suitable for cultivation in the cotton growing traits of Northern Zone and to explore the possibilities of introducing H4 and other hybrids in northern zone by studying its agronomy and plant protection measures.

Feteh was the first intra-hirsutum hybrid developed and released by PAU, Ludhiana for cultivation in the Punjab States in 1994. It was the first hybrid recommended in the northern India. At the same time the first intra arboreum hybrid LDH 11 was developed at PAU, Ludhiana in the Punjab State. Another intra-hirsutum hybrid LHH has been released for the Punjab State. This is the first cotton leaf curl virus resistant hybrid which has been released by PAU, Ludhiana in 1998.

14.3 Introduction of Bt-Cotton Hybrids

Bollworm complex is the major limiting factor to cause significant yield losses in cotton. Though Indian cotton area is first in the world but it is third in its production. Like many other parts of the world, the major reason for low productivity is damage caused by insect pests—notably by bollworm complex. In cotton crop, number of insecticides sprayed which valued a lot. Till date, none of the effective measures develop to manage Bollworm as at present Pink bollworm incidence is being noticed in the *Bt* cotton hybrid also. Hence, some variety should be develop which would be bollworm resistant. It can be done only through transgenics in the form of *Bt* cotton as there is no source of resistance to the bollworms. *Bt* cotton refers to transgenic cotton, which contains endotoxin protein inducing gene from soil bacterium *Bacillus thuringiensis*. The *Bt* cotton is of two types viz. bollgard which confers resistance to bollworms and roundup ready cotton which confers resistance to herbicides. Now there are two versions of bollgard i.e. the bollgard I contains Cry I Ac *Bt* gene in its genome while the bollgard II is the next generation transgenic Bt cotton which has combination of two genes viz. Cry 1 Ac and Cry 2 Ab into its genome and is more effective than bollgard I. Major advantages of *Bt* cotton include increase in yield, protection from bollworms, reduction in pesticide use etc.There are some limitations of *Bt* cotton which include high cost of seed mainly, effectiveness only upto 120 days, ineffective against sucking pests and promotes malpractices such as mixing of non-*Bt* seed with *Bt* seed and sale of F_2/F_3 seeds etc. Another major risk is that secondary pests such as mealy bug, whitefly and tobacco caterpillar were found to become economic pests due to less sprays on *Bt* cotton.

With the introduction of the *Bt* cotton hybrids, the farmers are more inclined to grow the *Bt* cotton hybrids in the South Western region of Punjab. Benefits of *Bt* technology are realized in farmer's community well, as it eradicates the need of spray against Bollworm pests (American Bollworm, Spotted Bollworm etc.) and allows uniform picking with considerable increase in yield. Initially, insecticide sprays were severely decreased against the Bollworm pest but it leads to the resurgence of the secondary pests (whitefly), which acts as the vector of CLCuD in cotton.

14.4 Cotton Leaf Curl Virus Disease–Earlier Reports

Cotton leaf curl virus disease (CLCuD) earlier known as African leaf curl of cotton was reported for the first time from Nigeria on *Gossypium peruvianum* and *G. vitifolia* by Farquharson in 1912. In India, CLCuD was first reported from Indian Agricultural Research Institute (IARI), New Delhi in 1989 and from farmers field in Sri Ganganagar, Rajasthan in 1993 (Ajmera, 1994; Varma et al., 1993) and Ferozpur district of Punjab adjoining to Pakistan border on *G. hirsutum* and afterwards it spread to entire North India in a short span of 4–5 years. The major constraint in cotton production now in North India is the cotton leaf curl virus disease, transmitted by the vector whitefly (Monga, 2014). Kranthi (2015) reported that there were two outbreaks of CLCuD in cotton in India, that is during 1993 and 1996.

14.5 Cotton Leaf Curl Virus Disease

Cotton leaf Curl Virus Disease (CLCuD), an important viral disease, caused by gemini virus belonging to family Gemini viridae, genus Begomovirus and transmitted by the vector whitefly (*Bemisia tabaci*). An infection at initial stages leads to drastic effect on the crop growth, yield, fibre quality etc. and CLCuD infected plants becomes stunted with twisted internodes. The disease was first noticed and initiated by small vein thickening (SVT) type symptoms on the lower sides of the young upper leaves of plants with netted like appearance. Later on, upward/downward leaf curling occurs. Leaves remain small, thickened and appears as a cup shaped, small leaflets (enations)/leaf-like outgrowths develop on the undersides of leaves on the main and lateral veins. Plant height, number of fruiting bodies and yield are drastically affected and reduced in the diseased plants.

14.6 Disease Rating Scale

The disease rating scale was finalized during AICCIP workshop and is being used with new modifications/suggestions till date for the screening or categorizing the plants against CLCuD (Monga, 2014). PDI of CLCuD in Cotton can be calculated by using the following formula:

$$\text{Per Cent Disease Index (PDI)} = \text{Average Grade}/\text{Maximum Grade} \times 100$$

**Disease index up to 30 with moderately resistant reaction will be permitted for varietal advancement or recommended to the farmers for the cultivation (Table 14.1).

14.7 Status of Cotton Leaf Curl Virus Disease (CLCUD) Virulent Strains in Northern India

As we all know that the (*Gossypium hirsutum* L.) hybrids and varieties are susceptible to the CLCuD and leads to the emergence of the disease complex symptoms i.e., an increase in the variants of the Cotton leaf curl Virus. The disease caused by one or more strains or new strains of existing virus or mixing of virulent strains caused the CLCuD disease complex, is the major factor for the frequent resistance breakdown among the *Bt* cotton hybrids and varieties against the viral diseases. Due to the combination of existing or new strains or some prevalent of some virulent

Table 14.1 Disease rating scale for cotton leaf curl virus disease

Disease severity (grade)	Symptoms	Disease index	Disease reaction
0	Complete absence of disease symptoms (immune)	0	Immune/ disease free
1	Symptoms of vein thickening on few upper leaves (small vein thickening)	0.1–10	HR
2	Small vein thickening + Main vein thickening + cupping and curling on few upper leaves	10.1–20	R
3	Top one-fourth of the plant affected with vein thickening, cupping and curling, leafy enations	20.1–30	MR
4	Half of the plant affected with vein thickening, cupping and curling, leafy enations	30.1–40	MS
5	Three-fourth of the plant affected with vein thickening, cupping and curling, leafy enations	40.1–50	S
6	Severely stunting of the plant with above symptoms	>50	HS

Source: Monga (2014)

strains of the CLCuD, variations in the severity of the CLCuD in the different locations of the cotton belt of the South Western Region of Punjab were noticed. At present, CLCuD-begomovirus species, Cotton leaf curl Multan Virus (CLCuMuV), Cotton leaf curl Kokhran Virus (CLCuKoV) and Cotton leaf curl Alabad Virus (CLCuAlV) have been identified in India (Ahuja et al., 2007; Rajagopalan et al., 2012; Kumar et al., 2010). Cotton leaf curl Rajasthan virus (CLCuRV) and Cotton leaf curl Burewala virus (CLCuBuV), were identified as predominant viruses in Cotton growing areas of North Western India.

14.8 Cotton Leaf Curl Virus Disease (CLCUD) and Its Drastic Impacts

CLCuD still being a serious limitation among other biotic and abiotic constraints for the cotton production. It was appeared as first epidemic during 1992–93 and declined the cotton yield up to 32% (Ali et al., 1995) and second epidemic was occurred during 2002–03 in Pakistan (Rajagopalan et al., 2012). The extent of losses vary with the degree of severity. The infection of CLCuD is depends upon the stage of crop, population of whitefly. More than 60% losses when the plant infection at seedling stage (Chopra et al., 1999).

The *Bt*-cotton hybrids now cultivated in about 94.75% area of the NW India are susceptible to this disease in the present condition (Monga et al., 2011.; Godara et al., 2015; Bhattacharyya et al., 2017). All the genotypes of *G. hirsutum* are susceptible to the CLCuD disease (100% infection) (Monga et al., 2011) whereas *G. arboreum* (Desi Cotton) are immune to the disease.

Arora and Singh (2016) conducted an experiment during the *Kharif* seasons of 2 succeeding years 2014–15 to determine the resistant source in cotton hybrids. They screened the 65 *Bt* cotton hybrids and 1 non-Bt cotton hybrid LHH 144 against the CLCuD. They examined all plants for the incidence and severity of CLCuD and collected the data within 30 days intervals up to 120 DAS of the crop. They revealed that out of the total 65 entries only the non-*Bt* LHH 144 showed highly resistant reaction towards CLCuD. Zubair et al. (2017) reported that CLCuD is a determinant problem in Indian subcontinent and it cause great economical losses to the cotton production.

14.9 Management of the Cotton Leaf Curl Virus Disease in Punjab

An integrated approach should be followed for the management of the Cotton leaf curl virus disease (CLCuD) and management strategy was mentioned in PoP of PAU,Ludhiana.

1. *Bt* cotton hybrids and varieties recommended by PAU, Ludhiana should be adopted for the cultivation in Punjab. Un-recommended ones must be avoided.
2. Sowing of the Cotton crop should be done latest by May 15, 2019 as the late sowing leads to aggravate the CLCuD severity.
3. *Desi* cotton varieties are immune to the CLCuD. *Desi* cotton varieties i.e. LD 1019, LD 949 and FDK 124 must be popularized among the farmers to enhance the area.
4. Cultivation of American cotton should be avoided around citrus orchards and adjoining to okra crops.
5. In early stages of the crop, CLCuD infected plants should be eradicated and destroyed from time to time as Eradication is the primary steps to check and widespread of the viral disease but it is not practically possible in cotton field if the large number of the plants are infected.
6. Clean cultivation in cotton crop should be adopted. Eradication of the weeds must be done in the fields and in the bund or areas adjoining the field. The weeds (i.e., *Kanghibuti* and *Peelibuti*) may act as the host reservoir of the vector whitefly, responsible for the transmission of CLCuD.
7. Volunteer ratoon cotton plants during off-season may be destroyed as it can be the reservoir host of the inoculums.
8. Judicious use of fertilizers especially, Nitrogen as the more application of Urea aggravates the attacks of insect pests (vector) of CLCuD.

If the attack of CLCuD is established then initially Neem based biopesticide (Nimbecidine or Achook) @ 1.0 l/acre or Homemade neem extract @1200 ml/acre will be selected for spray to manage the vector(whitefly) of CLCuD in initial stages and later on if the population of whitefly were enormous then go for the application of recommended chemical insecticides (Ulala 50 WG (Flonicamid), Osheen 20 SG (Dinotefuran), Lano 10 EC(Pyriproxyfen), Oberon/Voltage 22.9 SC (Spiromesifen), Dantotsu 50 WG (Clothianidine), Applaud 25 SC(Buprofezin) with appropriate dosages. It must be noted that for the spray of any insecticides, fix type solid cone nozzle must be used for the thorough coverage of plants and tank mixing as well as use of readymade insecticidal mixtures must be avoided as it may lead to resurgence of the vector (whitefly) of CLCuD in Cotton.

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