Effect of Cotton Leaf Mosaic Disease on Morphology, Yield and Fibre Characteristics of Upland Cotton in Pakistan

Khalid P. Akhtar, M. A. Haq, Wajid Ishaque, M. K. R. Khan and Azeem I. Khan

Nuclear Institute for Agriculture and Biology, P.O. Box-128, Jhang Road Faisalabad, Pakistan (Received on February 3, 2005; Accepted on February 23, 2005)

The effect of cotton leaf mosaic disease on morphology, vield and fibre characteristics was examined for a susceptible cotton candidate variety CRIS-168. Plants inoculated at most susceptible growth stage (six week) under screen house showed severe mosaic symptoms. There was a significant reduction in plant height and yield. Cotton leaf mosaic disease was found to produce severe effects on plant morphology with 24.1% reduction in plant height, 25% in internode length and 37.5% in number of sympodia on main stem. However no changes were observed against number of monopodial branches per plant. Inoculated plants showed 82% decrease in yield/plant, 80% in number of boll set/ plant, 12.1% in boll weight, 12.8% in lint weight, 10.8% in seed weight, and 6.8% in seed index. Cotton leaf mosaic disease also showed effects on fibre characteristics with 0.8% decrease in GOT and 1.6% in fibre length. In contrast, uniformity ratio, fibre fineness and maturity index was increased by 20.5%, 14.4% and 0.9%, respectively.

Keywords: cotton, leaf mosaic, fibre characteristics, Pakistan, plant morphology, yield components

Cotton (Gossypium hirsutum L.) is the most important fibre crop of Pakistan, and more than 60% of the total foreign exchange is earned through the export of raw cotton and cotton products. Cotton plant is infected by more than 75 fungal, bacterial and viral pathogens (Akhtar et al., 2001; Kirkpatrick et al., 2001). Many viral diseases of cotton are of economic importance worldwide, including cotton leaf crumple (CLCV) and cotton leaf curl virus (CLCuV) (Reddall et al., 2004). A new syndrome, named as cotton leaf mosaic was observed for the first time in Pakistan in 1994 at several locations in major cotton growing areas of Punjab (Ahmad et al., 2003). Infected plants show irregular mottle or yellow leaf mosaic, stunting, fewer flowers, boll shedding and reduced canopy. Symptoms on infected plant initiates with small chlorotic patches of discoloration,

ranging from 2-5 mm in diameter to more angular chlorotic patches ranging from 5-15 mm in diameter, which are the characteristic symptoms of cotton leaf mosaic disease. Veins of infected leaf become yellow, thickened and deformed. Symptoms are more apparent on young leaves, which can assume a much paler colour than healthy ones, with sterility and stunting of actively growing tips. Therefore, areas of infected cotton could be distinguished easily in the field even at a distance due to their light green appearance in contrast with the surrounding dark green healthy cotton (Akhtar et al., 2002; Brown 1992). The severity of the symptoms varies with the variety and environmental conditions (soil, climate and fertilizer) (Bink 1973; Brown 1992; Cauquil and Follin 1983; Fauquet and Thouvenel 1987). Based on ELISA, Tobacco Streak Virus (TSV) has been detected in samples showing mosaic symptoms (Ahmad et al., 2003; Nelson et al., 1998).

In Pakistan, cotton leaf mosaic is still considered to be a minor problem and extent of yield losses caused by this disease has not been established. This paper reports the first studies of cotton leaf mosaic disease effect on the morphology, yield and fibre quality of susceptible cotton variety CRIS-168.

Materials and methods

Source and maintenance of viral inoculum for graft inoculation. Cotton leaf mosaic disease inoculum was collected from naturally infected cotton plants of cotton variety CRIS-168 during 2002-03, exhibiting characteristic cotton mosaic symptoms. This isolate was maintained through grafting of infected stems (selected from the field) onto the plants of CRIS-168 grown in screen house.

Plant material. Seeds of variety CRIS-168 were sown in thirty pots (4-5 delinted seeds per pot) each placed in screen house. Thinning was done by maintaining one plant per pot two weeks after germination of seeds. Experimental material was maintained by the application of required fertilizer and plant protection measures. Clean tap water was used to irrigate the young seedling throughout the study.

^{*}Corresponding author.

Artificial inoculation. Fifteen plants of CRIS-168 were graft inoculated at the age of six week, with cotton leaf mosaic disease following the bottle shoot grafting method. For this purpose a single cut of 1-2 cm long and 0.1-0.2 cm deep was made on the stem near the 2nd last inter-node of each test plant. A 20-cm long cotton mosaic infected branch with growing tip was detached from the diseased plant (maintained culture). A similar cut (as on test plant) was made on this branch and corresponding cut surfaces were brought together and tied with parafilm. This stem was then placed in test tube (2-cm diameter and 16-cm length) containing distilled water. Distilled water was filled daily for seven days. After seven days tubes were removed and plants were monitored daily for symptom development. The data collected included grafting success, disease transmission, latent period (total time taken for first symptom appearance after grafting) and average disease severity 70 days post-grafting, using the disease rating scale as described by Bink (1973) given in Table 1.

Morphological characteristic. Individual plant height was measured in centimeters (cm) from the first cotyledonary node to the apical bud when the apical growth of the plant almost ceased. Then the number of sympodial branches on main stem and monopodial branches per plant were counted respective for diseased and healthy plants. Measurements for the internode length were made from three locations for each healthy and inoculated plant respectively. For this purpose first reading was made from third internode, 2nd from the middle of the plant and third from the apical node. All these measurements for each parameter were then averaged for the respective diseased and healthy plants.

Yield characters. At the end of the experiment mature bolls were harvested from each healthy and inoculated plant after counting green and mature bolls. Individual boll weights were summed, giving the total weight of mature bolls by each sample. Average boll weight was determined for each treatment (healthy and inoculated plant) by calculating the average of mature boll weight for all plants in a treatment. Using the mature boll weight values and the average boll weights of immature bolls remaining on the

plants at the end of the experiment, and assuming that all green bolls matured and contributed to the yield, a maximum harvest able yield (MHY) was estimated for each plant with the equation: MYH=MBW+(ABW × GB), where MBW is mature boll weight, ABW is average boll weight, and GB is green bolls (Brown et al., 1987). A seed index was also calculated by using the weight of 100 seeds for healthy and diseased plants.

Fibre characteristics. The seed cotton for each treatment (diseased and healthy plants) was ginned separately with a single roller electric gin. The lint thus obtained was weighed and ginning out turn percentage (GOT) was calculated by using the following formula:

Ginning out turn (%) =
$$\frac{\text{Weight of lint}}{\text{Weight of seed cotton}} \times 100$$

Fibre length and uniformity ratio was measured by high volume instrumentation (HVI 900), while fibre fineness and maturity index with the help of Sheffield micronaire instrument, according to standard test methods (ASTM 1997).

Results

Severe cotton leaf mosaic disease symptoms were found in all the graft-inoculated plants. The first disease symptom appeared as small chlorotic patches of discoloration on young leaves after 13-15 days (Table 2). Veins of infected leaves become yellowish thickened and deformed with sterility or fewer bolls and stunting of actively growing tips after 25 days of inoculation, similar to the symptoms observed in the field (Fig. 1 and 2).

Morphological characteristics. The effect of cotton leaf mosaic disease on plant morphology is shown in Table 3. Disease showed an adverse effect on the height of the inoculated plants. A decrease of 24.5% in the plant height (av. of 15 plants) of diseased plants was recorded as compared with those of healthy plants. A reduction of 37.5% in the number of sympodial branches on the main stem of the inoculated plants was also observed while no effects on monopodial branches was observed in case of

Table 1. Key for the assessment of cotton leaf mosaic disease severity

Category	Symptoms				
Type-1	Leaves exhibited small chlorotic patches of discoloration (ranging from 2-5 mm in diameter and vein yellowing).				
Type-2	Leaves showed large, more angular chlorotic patches ranging from 5-16 mm in diameter which can be considered the characteristic 'cotton mosaic' symptoms.				
Type-3	Symptoms involved chlorosis, vein thickening and deformation of leaf veins.				
Type-4	Stunting of the actively growing tips plus severe mosaic and sterility is observed.				

Table 2. Response of cotton plants of candidate variety CRIS-168 to cotton leaf mosaic disease through grafting under screen house conditions

Plant No.	Grafting success ^a	Disease transmission	Latent period	Disease severity after 70 days of grafting	Disease reaction
1	+	+	14	4	Highly susceptible
2	+	+	14	4	Highly susceptible
3	+	+	13	4	Highly susceptible
4	+	+	15	4	Highly susceptible
5	+	+	15	4	Highly susceptible
6	+	+	13	4	Highly susceptible
7	+	+	14	4	Highly susceptible
8	+	+	13	4	Highly susceptible
9	+	+	15	4	Highly susceptible
10	+	+	13	4	Highly susceptible
11	+	+	13	4	Highly susceptible
12	+	+	14	4	Highly susceptible
13	+	+	15	4	Highly susceptible
14	+	+	15	4	Highly susceptible
15	+	+	14	4	Highly susceptible
Average	100%	100%	14	4	Highly susceptible

^a Success of grafting was "+" when grafted stem (scion) survived and become the part of the root stock after the removal of test tube containing distilled water.



Fig. 1. Cotton plant showing cotton leaf mosaic disease symptoms after grafting.

inoculated plants as compared with healthy. **Seed cotton yield and yield components.** There was a drastic effect on the seed cotton yield of all the inoculated



Fig. 2. Cotton plant showing cotton leaf mosaic disease symptoms under field conditions.

plants, which showed a sharp decrease of 82.1%. Similarly number of bolls per plant and boll weight also showed a sharp decline of 80% and 12.1% respectively due to cotton leaf mosaic. In the diseased plants number of boll/plant ranged from 0 to 9. Besides cotton mosaic attack also brought about a marked reduction in lint weight, seed weight, and seed index as 12.8%, 10.8% and 6.8%, respectively as compared with healthy (Table 3).

Fibre characteristics. Negative effect of cotton leaf

Table 3. Effect of cotton leaf mosaic disease on morphology, yield and fibre characteristics of upland cotton variety CRIS-168

Sr. No.	Character	Healthy	Diseased	% decrease or increase over healthy
1	Boll wt. (g)	3.13	2.75	-12.1
2	Lint wt. (g)	1.17	1.02	-12.8
3	Seed wt. (g)	1.94	1.73	-10.8
4	Seed index	7.67	7.15	-6.8
5	GOT (%)	37.38	37.09	-0.8
6	Fibre length (mm)	26.53	26.40	-1.6
7	Uniformity ratio	39.47	47.56	+20.5
8	Fibre fineness (μg/inch)	4.24	4.85	+14.4
9	Maturity index	87.8	88.58	+0.9
10	Plant height (cm)	153.4	115.8	-24.5
11	No. of bolls / plant	25.0	5.0	-80.0
12	No. of monopodia / plant	4.0	4.0	0.0
13	No. of sympodia / plant	16.0	10.0	-37.5
14	Internode length (cm)	5.56	4.17	-25.0
15	Yield / plant (kg)	0.078	0.014	-82.1

mosaic disease was also observed on GOT% and staple length of the inoculated plants with over all slight reduction of 0.8% and 0.7%, respectively as compared to healthy. However data depicted that the cotton leaf mosaic disease has adverse effects on fibre fineness, maturity index and uniformity ratio. There was a slight increase of about 0.9% in maturity index and significant increase of about 14.4% and 20.5%, respectively in fibre fineness and uniformity ratio in fibre from infected plants over healthy plants (Table 3).

Discussion

Symptoms of the cotton leaf mosaic disease were found in all the fifteen inoculated plants with disease severity category type-4 (Table 2). The relative high disease severity from early inoculation and the fact that the above ground yield components were significantly reduced, confirm that the disease can cause considerable production losses. Symptoms exhibited by the inoculated plants under screen house resembles those reported elsewhere (Bink 1973; Brown 1992; Cauquil and Follin 1983; Mahmood and Tahir 2001) and include plants stunting, severe mosaic, vein thickening and deformation of leaves & veins of leaves with sterility.

It was apparently from this study that cotton leaf mosaic disease reduced plant height and number of sympodial branches on main stem and internode length, which constitute the major above ground yield components. Viral infection also reduced the number of bolls (80%) per plants. In Africa cotton mosaic caused huge losses in yield in cotton variety BJA-592 (Brown, 1992). All the inoculated plants were stunted with severe mosaic symptoms, impact

on the overall seed cotton yield of cotton is likely to be enormous. Indeed infected plants produced only a few bolls or sterile as compared to the healthy ones and this decrease with the increase in disease severity. The highest reduction in the above ground yield components was recorded in the terms of yield/plant followed by number of boll sets per plant (Table 3) and it points out the threat that cotton leaf mosaic infection can pose on the yield of seed cotton. There was significant reduction in number of sympodia/plant, internode length and plant height while number of monopodial branches per plant were not effected.

Second most important aspect of the current study was to assess the effects of cotton leaf mosaic disease on the fibre characteristics. Cotton leaf mosaic caused minor reduction in the GOT and fibre length. However, there was slight increase in fibre uniformity and fineness. This study corresponds to early findings of Ahmad et al. (2002) and Mahmood et al. (1996) which reported the same trend against CLCuV disease.

At present CLCuV is the major constraint of cotton production in Pakistan. Despite the fact that cotton leaf mosaic disease is still considered to be a minor problem in Pakistan, some interesting results on varietal susceptibility is now available. Varieties like CIM-434, CIM-435, CIM-443, CIM-445, CIM-448, CIM-1100, LRA-5166, BH-100, FH-634, VH-53 and VH-55, which are resistant against CLCuV are susceptible to cotton leaf Mosaic (Ahmad et al. 2003). In contrast varieties like CIM-70, S-12, B-622, B-30, B-496, BH-4, BH-89, BH-95 and NIAB-Krishma are susceptible to CLCuV while resistant to cotton leaf mosaic disease (Ahmad et al., 2003). In addition a number of other varieties are susceptible to both diseases (Ahmad et al., 2003; Nelson, 1998). This study has revealed significant

reduction in above ground yield components, which suggests that cotton leaf mosaic can be a very serious disease and may cause substantial losses. Present investigations give an idea that cotton breeders, who are actively engaged in evolving resistant varieties against CLCuV, may also consider this disease and adopt suitable strategies to tackle the problems of cotton leaf curl virus and cotton mosaic at the same time.

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