REVIEW

ADOPTION OF BT COTTON: THREATS AND CHALLENGES

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Adopting new technology always involves advantages and risks; Bt cotton (*Gossypium hirsutum* L.) is a new technology well known in developed countries for its many advantages, such as reduced pesticide application, better insect pest control, and higher lint yield. However, its success in developing countries is still a question mark. Global adoption of Bt cotton has risen dramatically from 0.76 million ha when introduced in 1996 to 7.85 million ha in the 2005 cotton-growing season where 54% of the cotton crops in the USA, 76% in China, and 80% in Australia were grown with single or multiple Bt genes. Bollworms are serious cotton pests causing 30-40% yield reduction in Pakistan and 20-66% potential crop losses in India. The major advances shown in this review include: (1) Evolution of Bt cotton may prove to be a green revolution to enhance cotton yield; (2) adoption of Bt cotton by farmers is increasing due to its beneficial environmental effects by reducing pesticide application: however, a high seed price has compelled farmers to use illegal non-approved Bt causing huge damage to crops because of low tolerance to insect pests; and (3) some factors responsible for changes in the efficiency of the Bt gene and Bt cotton yield include internal phenology (genetics), atmospheric changes (CO_2 concentration), nutrition, insect pests, boll distribution pattern, disease and nematodes, removal of fruiting branch and/or floral bud, introduction of Bt gene, and terpenoids and tannin production in the plant body.

Key words: Bt cotton, management, *Gossypium hirsutum*.

Every new technology has its benefits and risks; the benefits associated with using transgenic crops are a dramatic decrease in the use of conventional and broad-spectrum insecticides and target pests, yield improvement, lower production costs, and compatibility compared with other biological control agents (Arshad *et al*., 2007). Risks include out-crossing by pollen transfer to non-transgenic plants, food safety concerns, development of resistance in target pests, and effects on non-target organisms and biodiversity (Cannon, 2000; Wolfenbarger and Phifer, 2000; Edge *et al*., 2001; Shelton *et al*., 2002; Naranjo, 2005).

During the 1980s, genetic engineering of crops was first accomplished by Fischoff *et al*. (1987), who inserted genes to produce an insecticidal endotoxin from *Bacillus thuringiensis* (Bt) bacterium into tomato and tobacco plants. *Bacillus thuringiensis* genes inserted into cotton (*Gossypium hirsutum* L.) 'Kurstaki' produced the *Cry 1 Ac* protein that was especially toxic to the lepidopteran insect species (Perlak *et al*., 1990). Insecticides employed in cotton against the bollworm complex were 50% of the total insecticide volume used in agriculture (Fitt, 2008). Bt varieties globally reduced the insecticide active ingredient (ai) applied by 19% (Brookes and Barfoot, 2006). The revolution in cotton production on a global scale is due to the cultivation of transgenically modified cotton that expresses insecticidal proteins derived from *B. thuringiensis* (Head *et al*., 2005). In 1998, Bt cotton boosted total USA lint production by 38.6×10^6 kg (Gianessi and Carpenter, 1999). In China, Bt cotton reduced total insecticide use by 60-80% compared with conventional cotton in 1998 (Xia *et al*., 1999). In the USA, more than 84% of Bt cotton growers were satisfied with it, while more than 73% of Bt cotton users indicated that they are more satisfied with Bt cotton than conventional cotton cultivars (Marketing Horizons, 1999). Insect-resistant Bt cotton is rapidly dominating world cotton production (Jenkins *et al*., 1995; Pray *et al*., 2002). The first Bt transgenic cotton variety (called Bt cotton), which expressed the same gene construct of *Cry I Ac*, was commercially released in Australia (Ingard™ cotton) and in USA (Bollgard™ cotton) in 1996 (Olsen and Daly, 2000). Cotton is Pakistan's main cash crop and is known as "White Gold" (Arshad *et al*., 2007). Pakistan is the fourth largest cotton producer (Abro *et al*., 2004) after China, the USA, and India.

Bt cotton use by farmers in Pakistan increased in 2010.

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In Punjab and Sindh, almost 80% of the area grows Bt cotton (Australian Bt) with a high incidence (60-100%) of *Cotton leaf curl virus*. In 2010, the Pakistan Agricultural Research Council (PARC) imported almost 950 kg of five different Bt cotton seed varieties from China with special permission to conduct direct trails in farmers' fields without following the rules and regulations designed by the National Biosafety Committee (NBC), Pakistan Central Cotton Committee (PCCC), and the Federal Seed Certification and Registration Department (FSC&RD) (Government of Pakistan, 2010). Approximately 22% of the global cotton area was planted with Bt cotton in 2003; two major cotton producing countries were USA and China with 48% and 57%, respectively (James, 2003). The global area covered by genetically modified (GM) crops in 2009 and 2010 is shown in Table 1.

Adopting Bt cotton

Bt cotton expressing the *Cry I Ac* toxin derived from *B. thuringiensis* was first commercialized in the USA in 1996; it successfully controlled lepidopteran pests, especially bollworms which are the main constraint in cotton productivity (Brookes and Barfoot, 2006). Global adoption of Bt cotton has risen dramatically from 0.76 million ha when introduced in 1996 to 7.85 million ha in 2005. It is remarkable that 54% of cotton crops in the USA, 76% in China, and 80% in Australia were grown with single or multiple Bt genes in 2005 (Arshad *et al*., 2007). Worldwide, GM cotton that included Bt and the dual-stacked herbicide-tolerant Bt gene was planted in over 15.5 million ha in 2008 and constituted approximately 43% of the cotton area (Gujar *et al*., 2007). In China, Bt cotton has spread quickly in the provinces where initial approval was given along with impact studies (Pray *et al*., 2002; Table 2). Field studies in China have shown that farmers have reduced pesticide and labor costs by adopting Bt cotton; moreover, there is less exposure to toxic insecticides (Xia *et al*., 1999; Pray *et al*., 2002). In Pakistan, eight Bt varieties were approved for field trails in 2009; few studies have attempted to make a preliminary performance comparison of existing Bt varieties with recommended non-Bt varieties (Arshad *et al*., 2009). These studies observed a relatively poor performance of existing Bt cotton compared with recommended conventional varieties; PARC found that these varieties produced less toxin protein (PARC, 2008). Many studies have analyzed the impact of Bt cotton in developing countries (Thirtle *et al*., 2003 for South Africa; Hue *et al*., 2002 for China; Gandhi and Namboodiri, 2006 for India), which suggest a decline in pest infestation, higher yield, and higher profit after adopting Bt cotton (Table 3). Table 3 compares adopters and non-adopters of Bt cotton. Data sources included farm surveys and on-farm experimental plots (India). Bt technology will be the major factor in boosting agricultural productivity, especially in developing countries with additional positive effects on human health and the environment due to reduced pesticide levels (Pemsl *et al*., 2004). Qaim and de Janvry (2003) opined that the high technology price of Bt seed inhibits its adoption.

Table 1. Global distribution of genetically modified (GM) crops in 2009 and 2010.

Rank	Country	2010 Area (million ha)	2009 Area (million ha)	Biotech crops
1	USA	66.8	64.0	Soybean, maize, cotton, canola, squash, papaya, alfalfa, sugarbeet
2	Brazil	25.4	21.4	Soybean, maize, cotton
3	Argentina	22.9	21.3	Soybean, maize, cotton
4	India	9.4	8.4	Cotton
5	Canada	8.8	8.2	Maize, soybean, canola, sugarbeet
6	China	3.5	3.7	Cotton, tomato, poplar, papaya, sweet pepper
7	Pakistan	2.4		Cotton
8	South Africa	2.2	2.1	Soybean, maize, cotton

Source: James, 2010.

Factors affecting quality, yield, and yield components of Bt cotton

Genetics. Extensive studies comparing transgenic cotton varieties with their recurrent parents showed that fiber uniformity, length, strength, and elongation showed no significant differences due to transgenic technology (Ethridge and Hequet, 2000). Cooke *et al*. (2001) compared commercial yields and quality reports of cotton varieties from 12 to 15 Mississippi Delta Farms for the 1997-2000 period. Data were taken to measure the entomological and economic impact of Bt cotton compared with conventional cotton. There were no significant differences in staple length and grade between transgenic and conventional varieties observed on all the

Table 2. Performance difference (% change) between Bt and conventional cotton varieties.

	Argentina	China	India	Mexico	South Africa
Bt cotton[1], % of cotton area	5	40	2	71	10
Lint yield, % change	33	19	80	11	65
Chemical sprays, N°	-2.4	-13.2[2]	-3.0	-2.20	na
Pest control costs, % change	-47	-67	-39	-77	-58
Seed cost, % change	530	95	82	165	89
Profit, % change	31	340	83	12	299

na: Non available.
Source: Argentina: Qaim and de Janvry, 2003; China: Pray *et al*., 2002; India: Qaim and Zilberman, 2003; Mexico: Traxler *et al*., 2003; South Africa: Bennett *et al*., 2003.
Data are means of all surveyed years.
[1]James, 2003; FAOSTAT, 2004.
[2]1999 data.

Table 3. Performance of Bt cotton as regards insecticide reduction, increase in yield, and gross margin.

Country	Insecticide reduction	Increase in effective yield	Increase in gross margin	References
	%	%	US\$ ha^{-1}	
Argentina	47	33	23	Qaim and de Janvry, 2005
Australia	48	0	66	Fitt, 2003
China	65	24	470	Pray *et al*., 2002
India	41	37	135	Subramanian and Qaim, 2009
Mexico	77	9	295	Traxler *et al*., 2003
USA	36	10	58	Carpenter *et al*., 2002

farms over the 4 yr. Creech (2001) compared yields and fiber quality data from 20 conventional varieties and 20 transgenic varieties; results showed that conventional varieties exhibited slight advantages in mean length and uniformity and transgenic varieties were slightly better (lower) in micronaire. Tested conventional varieties had approximately 4% higher strength.

Environmental conditions. Ongoing changes in textile processing, particularly spinning technologies, have led to increased emphasis on breeding for both improved yield and fiber quality (Patil and Singh, 1995). Studies of gene action and heterosis have suggested that there is little non-additive gene action in fiber length, strength, and fineness in cotton genotypes (Meredith and Bridge, 1972). Sizable interactions between combined annual environments and fiber strength have suggested that environmental variability can prevent the full realization of genotype fiber-quality potential (Green and Culp, 1990). However, early (pre-1980) statistical comparisons of the relative genetic and environmental influences on fiber strength suggest that they are conditioned by only a few major genes (May, 1999).

Nutrition. Better fertilization at an early stage could substantially improve Bt cotton yield (Hofs *et al*., 2006b). Nitrogen application at a rate of 50 kg ha^{-1} increased seed protein content (Patil *et al*., 1997). Total N contents of Bt cotton cultivars were significantly higher than their parents during the peak square and boll period. Leaf N uptake of Bt cotton increased after introducing the Bt gene (Chen *et al*., 2005b). Excesses of N delay maturity, promote vegetative tendencies, and usually result in lower yields (McConnell *et al*., 1996). Averaged over the years, the number of opened bolls per plant was significantly greater at 143 kg N ha^{-1} than 95 kg N ha^{-1} (Sawan *et al*., 2006). Nitrogen deficiency has been observed to decrease auxin content and markedly increase inhibitor content in leaves and stems (Anisimov and Bulatova, 1982). Boll weight increased as the N rate increased from 95 to 143 kg ha^{-1} (Sawan *et al*., 2006). The increase in boll weight can be due to an N-induced increase in mineral uptake and to photosynthate assimilation and accumulation in sinks (Breitenbeck and Boquet, 1993). Nitrogen fertilizer increased leaf photosynthetic rates by 11-29% when plants were given up to 157 kg N ha^{-1} (Cadena and Cothren, 1995). The increase in seed index can be due to enhanced photosynthetic activity since N is an essential component of chlorophyll (Bondada and Oosterhuis, 2000). Nitrogen plays the most important role in building the protein structure (Frink *et al*., 1999). Seed development requires both N and C skeletons (Patil *et al*., 1996). Nitrogen deficiency produced ethylene at an early crop development stage, which resulted in increased square, flower, and boll shedding in cotton (Legé *et al*., 1997).

As Bt cotton varieties are adopted and yield per unit area continues to increase, there is a rising frequency of K deficiency in many cotton-growing countries. Potassium deficiency decreases leaf area index, photosynthesis, and plant biomass but enhances earliness of maturity (Hezhong *et al*., 2004). Potassium deficiencies in cotton have become more common, particularly in modern high-yielding cotton varieties such as Bt transgenic cotton (Phipps *et al*., 2003). Additional K resulted in more aboveground biomass partitioned to vegetative parts (Gwathmey, 2005). Potassium deficiency slightly increased the proportion of DM in reproductive organs by cutout, although total aboveground dry weight was not affected by K (Pettigrew *et al*., 2005). Gwathmey and Howard (1998) also observed that additional K delayed maturity, but their study only compared deficient and adequate K rates Pettigrew *et al*. (2005) concluded that K deficiency resulted in earlier cotton crop maturity. Potassium plays a key role in assimilation, long distance assimilate transport, phloem loading, N metabolism, storage process, osmotically active cation, control of water relationship in plants, response of crops to adverse climatic and soil conditions, and plant resistance and tolerance to pathogens (Hezhong *et al*., 2004). Potassium is an essential nutrient for the reproductive development of cotton, partly due to its role in carbohydrate transport in developing bolls. Potassium deficiency reduced translocation of photoassimilates to bolls (Ashley and Goodson, 1972), resulting in decreased lint yields (Pettigrew, 1999). In the cotton crop, as the season progresses, premature senescence symptoms can spread and the crop is defoliated. This K-related premature senescence was initially found in many countries, including Australia (Wright, 1999), China (Zheng and Dai, 2000), and the USA (Oosterhuis, 2001). Adequate K may also be needed to efficiently use N fertilizer (Varco and Fridgen, 2004). Potassium probably exerts its greatest effects on disease through specific metabolic functions that alter compatibility relationships of the host-parasite environment (Kafkafi et al., 2001).

Insect pests. Environmental adversities limit photosynthate availability to the developing organs and lead to the shedding of fruiting forms (Guinn, 1985); additional loss due to entomological factors causes shedding of fruiting forms by 75-80% in rain-fed cotton (Bhatt *et al*., 1972). Bt cotton has been commercialized to protect the losses of fruiting forms by the entomological factors because Bt cotton has better retention of early-formed squares and bolls due to better insect control. Bt plants were full of developing bolls on the lower canopy, while non-Bt plants had few squares, flowers, and developing bolls spread intermittently on the canopy (Hebbar *et al*., 2007). This resulted in yield improvement with Bt cotton cultivation as shown by earlier studies (Qaim and Zilberman, 2003).

Bt cotton varieties incorporate the Bt *Cry I Ac* gene and show resistance to certain insect pests (Perlak *et al*., 2001). Transgenic cotton cultivars showed their possible role in controlling three main pests, that is, *Helicoverpa armigera*, *Diparopsis castanea,* and *Earias biplaga* (Green *et al*., 2003). Growing Bt cotton has become a key measure to effectively control damage caused by the cotton bollworm *Pectinophora gossypiella* and *H. armigera* (Wu *et al*., 2003; Li *et al*., 2006). With the commercial cultivation of Bt cotton, the infestation of both pink and cotton bollworms tends to gradually decrease, while the risk of severe damage in certain regions significantly diminishes (Wu and Guo, 2005). Decreasing insecticide application in Bt cotton fields increased the diversity of beneficial natural enemies such as ladybugs *Chrysopa* spp., while spiders effectively controlled the development of harmful insect populations in cotton (Wu *et al*., 2003).

In contrast to conventional cotton, Bt cotton has the potential to impact ecological environments by possible extensive planting of Bt-transgenic plants. Large-scale planting of Bt cotton mainly involves changes in the secondary pest populations of target insects such as aphids, spider mites, and mirids; this can lead to new problems in cotton pest control when these minor pests can become the major pest (Wu, 2007). The toxic effects of Bt cotton on lepidopteran insects can affect the food chain in the agro-ecosystem and lead to an imbalance of the ecosystem; the occurrence of resistant target pests can lead to the ineffectiveness of Bt cotton (Wu and Guo, 2005). The control efficacy of Bt cotton on different insect pests is shown in Table 4.

Disease and nematodes. Disease and nematode pathogens negatively impact yield and cause severe losses. Major bacterial and fungal pathogens include seedling diseases (e.g., *Rhizoctonia solani*, *Fusarium oxysporum*, *F. solani*, *Pythium ultimum*, *Thielaviopsis basicola*, and other *Pythium* and *Fusarium* species), fungal wilt diseases (e.g., Verticilium and Fusarium wilt), root rots (e.g., *Phymatotrichum omnivorum*, *T. basicola*, and *Pythium* species), and foliar diseases (e.g., *Xanthomonas campestris* pv. *malvacearum*) (Bell, 1999). The most important nematode pathogens are *Meloidogyne incognita* (root-knot nematode) and *Rotylenchulus reniformis* (common reniform nematode) (Robinson, 1999). Therefore, characters should also be induced during the development of Bt cotton disease.

Table 4. Control of Bt cotton on different insect pests.

Insect pests	Bt cotton control	References
Pectinophora gossypiella	95%	Wu and Guo (2005)
Spodoptera litura	13.3-53.3%	Deng *et al*. (2003)
Heliothis virescens	96%	Henneberry *et al*. (2001)
Spodoptera exigua	57%	Henneberry *et al*. (2001)
Heliothis virescens	95%	Moore *et al*. (1997)

Boll distribution pattern. The boll distribution pattern can explain the origin of yield differences by assessing pest damage and crop management in the field (Kerby and Bruxton, 1981). Hofs *et al*. (2006a) recorded boll distribution on the plant under optimal large-scale irrigated farming conditions; the transgenic variety was found to have undisputed advantages by providing better early plant protection, earlier picking, and nearly 13% higher yields than the non-Bt cotton variety. Better boll retention on the first fruiting branch is an agronomic advantage (Constable, 1991) and varieties that are able to keep their fruits in the first position improve production earliness (Ungar *et al*., 1987). Sahai and Rahman (2003) recorded Bt plants with less vigorous growth, fewer branches, smaller leaves, and smaller bolls than non-Bt cotton cultivars, while Dong *et al*. (2006) observed increased growth and yield of Bt plants compared with conventional cultivars. Photosynthesis and growth during boll development are positively associated with boll load in Bt cotton, and the higher sink activity lowered Bt source to sink ratio leading to faster senescence and crop maturity than non-Bt cotton cultivars (Hebbar *et al*., 2007).

Removal of early fruiting branches and/or floral bud. Removing fruiting forms usually enhances vegetative growth and development, such as increased plant height, total dry weight, leaf area index (LAI), and lengthening of anthesis (Ungar *et al*., 1987; Jones *et al*., 1996). Removing early-season flower buds increased root growth, while removing fruiting branches alters spatial yield distribution (Sadras, 1995; Bednarz and Roberts, 2001; Dumka *et al*., 2004). Removing early flower buds can also increase single leaf as well as canopy photosynthesis rate (Jasoni *et al*., 2000; Wells, 2001; Dumka *et al*., 2003), which has been considered one of the most important mechanisms for plant growth and yield compensation in cotton. Removing early fruiting branches can reduce premature senescence through decreasing the sink/source ratio, and thus enhance Bt transgenic cotton cultivar yield and quality (Dong *et al*., 2008). Removing early fruiting branches significantly altered the sink/source ratio through the delayed initiation of fruiting and enhanced vegetative growth, plant height, leaf area, yield, and fiber quality, such as fiber strength and micronaire in Bt cotton (Dong and Li, 2007).

Bt gene on nitrogen metabolism. Introducing the Bt gene and expressing the insecticidal protein content have caused alterations in metabolic processes related to both vegetative and reproductive growth (Tian *et al*., 2000; Chen *et al*., 2002). Alterations in vegetative and reproductive growth affected the expression of lint potential and fiber quality (Chen *et al*., 2005a). It led to increased plant height (Kerby *et al*., 1995), higher relative growth rate and biomass (Godoy *et al*., 1998), smaller bolls (Tian *et al*., 2000), and reduced fiber micronaire and lint percentage (Kerby *et al*., 1995; Fu *et al*., 2001). Leaf insecticidal protein content of Bt cotton was closely correlated with glutamic-pyruvic transaminase (GPT), nitrate reductase (NR), and protease activity (Chen *et al*., 2003), thus indicating that the expression of the external Bt gene affected Bt cotton N metabolism; the changed growth characteristics could be related to Bt cotton N metabolism (Chen *et al*., 2005b).

Wang *et al*. (1998) reported that Bt transgenic cotton lines increased leaf amino acid content, but more nutrients were used for stem and branch growth. Breen *et al*. (1999) reported that high N resulted in more vegetative growth; therefore, significantly less lint was produced in the Bt cotton cultivar. Similarly, Dong *et al*., (2000) reported that an increase of leaf NR activity and NO_3-N enhanced boll shedding for Bt cotton cultivars. Bt cotton 'CCRI-30' had smaller bolls than its parent because of the poor supply of assimilates (Tian *et al*., 2000). Jackson and Gerik (1990) reported that N deficiency in the boll caused a decline in boll size and an increase in boll shedding; further amino acid metabolism affected boll development of linted ('Suvin', 'MCU 5') and lintless ('MCU 5' mutant) cotton genotypes (Perumal and Naidu, 1987). Deotale *et al*. (1988) also reported that vegetative selection against high levels of amino acids and the boll from squaring until boll maturation can be used as an index in breeding resistance to boll shedding. Total N reduced sharply in the bolls of Bt cotton cultivars and reducing total N decreased N metabolism and limited boll development; there was a significantly positive correlation between GA3 content at flowering and boll size at 10 and 20 d after anthesis, respectively ($r = 0.99^*, 0.96^*$) in Bt cotton cultivars (Chen *et al*., 2005a). This result also suggests that reducing GA_3 can induce declining N absorption and metabolism, thus affecting boll development (Kishor and Mehta, 1987).

Toxicity of Bt cotton. Both environmental and genetic factors have been proposed to help explain the variation in toxicity of Bt cotton, including cultivar background and site-of-gene insertion (Sachs *et al*., 1998). A decreased expression of the *Cry I Ac* gene (Finnegan *et al*., 1998) reduced the amount of *Cry 1 Ac* protein (Holt, 1998). Plant effects could include metabolic changes in the plant in response to growth and reproduction (Benedict *et al*., 1996). Environmental factors include time of planting, location (Fitt, 1998), and N or water availability (Benedict *et al*., 1996). Benedict *et al*. (1996) used enzyme-linked immunosorbent assay (ELISA) to quantify *Cry 1 Ab* and *Cry 1 Ac* levels, but they noted that the yield of extracted protein was less as the plant matured. Holt (1998) developed ELISA methods to quantify *Cry 1 Ac* protein in Australian cotton. She noted a decline in *Cry 1 Ac* levels that correlated ($r^2 = 0.83$) with the increased survival of *H. armigera* first-instar (Fitt, 1998).

Cry 1 Ac protein content in Bt cotton was significantly reduced by high temperatures (Chen *et al*., 2005a), NaCl stress (Jiang *et al*., 2006), and N deficiency (Coviella *et al*., 2002), whereas others show that a high N fertilizer rate (Pettigrew and Adamczyk, 2006) or foliar applications of the plant growth regulator Chaperone greatly improved *Cry 1 Ac* protein levels, thus resulting in increased mortality of neonate bollworms feeding on treated plants (Oosterhuis and Brown, 2004).

Effect of terpenoids and tannins on efficacy of Bt protein

Field observations of Australian Bt cotton showed that plants expressing the *Cry 1 Ac* protein are less toxic to *H. armigera* first-instar when leaves are from fruiting versus pre-square plants (Olsen and Daly, 2000). Terpenoids fluctuate temporally (Zummo *et al*., 1984) and condensed tannin levels generally increase with plant age (Lege *et al*., 1992), so that both can play some part in changing the efficacy of *Cry 1 Ac* protein. There are few reports on the interactions between Bt proteins and terpenoids, although Sachs *et al*. (1996) found that they enhanced the efficacy of transgenic Bt cotton against *H. virescens*.

Tannins can alter the efficacy of Bt toxins against target species. Arteel and Lindroth (1992), Sivamani *et al*. (1992), Gibson *et al*. (1995), and Morris *et al*. (1995) all reported increased mortality in lepidopteran species when hydrolysable tannin compounds were combined with various Bt toxins in bioassays.

In summary, the variation in toxicity of Bt cotton between the pre-square and fruiting stages not only resulted from changes in the concentration of *Cry 1 Ac* protein but also plant-toxin interactions that altered *Cry 1 Ac* protein toxicity or availability (Olsen and Daly, 2000).

Effects on Bt protein of removing early fruiting branches

Removing early fruiting branches in Bt cotton increased lint yield (5.2-7.5%) and boll size (5.1-5.7%), and a higher level of *Cry I Ac* protein was found in fully-expanded young leaves in removed fruiting branches compared with the control plant; this clearly indicated that removing fruiting branches enhanced *Cry 1 Ac* expression (Dong *et al*., 2008). Removing fruiting forms leads to great morphological and physiological changes, including lint yield variation ranging from a small increase to a large decrease (Sadras, 1995). Nitrogen metabolism affected *Cry 1 Ac* protein content (Chen *et al*., 2005b), and removing early fruiting forms could change N metabolism (Deng *et al*., 1991); therefore, removing early fruiting branches can increase *Cry 1 Ac* protein content in Bt cotton plants.

Effect of elevated CO_2 on Bt cotton performance

The concentration of atmospheric CO_2 has risen from 280 to 360 ppm because of the industrial revolution and this level is anticipated to double by the end of this century (Houghton *et al*., 2001). The increase in atmospheric concentration can have a variety of direct and indirect effects on relationships between host plant, their herbivores, and the herbivores' natural enemies (Stiling *et al*., 2002). Elevated CO_2 tends to increase photosynthetic rates, growth, yield, and C:N ratio in most C_3 plants (Cure and Aycock, 1986; Bazzaz, 1990). Only limited research has been reported about the effect of elevated CO_2 on transgenic Bt cotton or the effects on bollworms fed Bt cotton grown in elevated CO_2 (Coviella *et al*., 2002). Elevating the CO_2 level from 330 to 660 ppm has led to a 95% yield increase in cotton (Kimball, 1986). Increases in soluble sugar, starch, total non-structural carbohydrates (TNC), TNC:N ratio, condensed tannin, gossypol, and decreases in water content, N, and Bt toxin protein were observed in young bolls from cotton plants grown under elevated CO_2 conditions compared with those in ambient CO_2-grown cotton for both Bt and non-Bt cotton (Chen *et al*., 2005a). The most herbivorous insects appear to be negatively affected by elevated CO_2 because foliar N decreased and the C:N ratio increased, except for phloem-feeding insects (Watt *et al*., 1995; Bezemer and Jones, 1998). Nitrogen contents limit insect growth and development because N is the single most important limiting resource for phytophagous insects (Mattson, 1980). Bt cotton delayed the larval life cycle, reduced body weight and fecundity, and significantly reduced larval RGR and MRGR. In contrast, elevated CO_2 did not significantly affect growth and development of cotton bollworms compared with the cotton variety; however, effects of transgenic Bt cotton on growth and development of cotton bollworms were enhanced when grown under elevated CO_2 conditions (Chen *et al*., 2005a).

CONCLUSIONS

Cotton fiber is a more challengeable fiber than synthetic fiber; it is inevitable that the per unit yield of cotton will increase to fulfill the basic human need for clothing. Bt technology is one of the best approaches in developed countries. Bt cotton should perform equally well in countries like Pakistan. Although the primary controlling factor is availability of a true type seed, there is an urgent need to develop new package production technology for Bt cultivars and also create awareness about it among farmers. For example, while using Bt cotton, farmers do not use insecticide even against sucking pests so that minor pests become major ones causing a hidden yield reduction in Bt cotton. Similarly, a balanced and timely application of N, K, and other macro- and micronutrients is important to obtain a higher Bt cotton yield. Moreover, Bt cultivars are more sensitive to heat stress, so their performance must be investigated under changing climates, especially under increased CO_2 concentration.

Adopción de algodón Bt: desafíos y amenazas. La adopción de nueva tecnología siempre involucra ventajas y riesgos; algodón Bt (*Gossypium hirsutum* L.) es una nueva tecnología bien conocida en países desarrollados por muchas ventajas como reducida aplicación de pesticidas, mejor control de insectos plaga, y mayor producción de fibra, pero su éxito en países en desarrollo aún conlleva dudas. La adopción global de algodón Bt ha aumentado dramáticamente de 0,76 millones de hectáreas en su introducción en 1996 a 7,85 millones de hectáreas en la estación de cultivo de algodón 2005, 54% de cultivos de algodón en EE.UU., 76% en China, y 80% en Australia se cultivaron con genes Bt únicos o múltiples. Los gusanos del algodonero son plagas graves del algodón que causan 30-40% de reducción en rendimiento en Paquistán, y 20-60% pérdidas potenciales en India. Las principales ventajas mostradas en esta revisión incluyen: (1) Evolución de algodón Bt puede probar revolución verde en el aumento de rendimiento de algodón; (2) adopción de algodón Bt por agricultores está aumentando debido a sus efectos beneficiosos en el medioambiente al reducir aplicación de pesticidas, pero el alto precio de la semilla ha obligado a los agricultores a usar semilla Bt ilegal no aprobada que causó gran daño al cultivo debido a baja tolerancia a insectos plaga; y (3) algunos factores responsables de cambios en eficiencia de genes Bt y por lo tanto de rendimiento del algodón Bt incluyen fenología interna (genética), cambios atmosféricos (concentración de CO_2), nutrición, insectos plaga, patrón de distribución de gusanos, enfermedades y nematodos, remoción de ramas fructificantes y/o yemas florales, introducción de genes Bt y producción de terpenoides, taninos, etc., dentro del cuerpo de la planta.

Palabras clave: algodón Bt, manejo, *Gossypium hirsutum*.

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