SILVER JUBILEE LECTURE SERIES

GENETICALLY MODIFIED CROPS IN INDIA WITH SPECIAL REFERENCE TO COTTON

BY

Dr. P. K. GHOSH

Advisor, Department of Biotechnology,
Ministry of Science and Technology,
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JUNE 23, 2000

Indian Society for Cotton Improvement
C/o Central Institute for Research on Cotton Technology
Matunga, Mumbai 400 019

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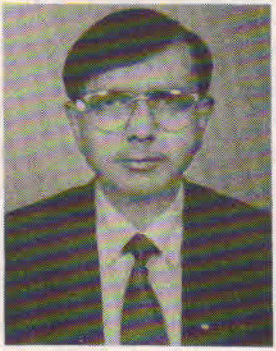

Dr. P. K. GHOSH

Dr. P.K.Ghosh did his M. Tech in Chemical Engineering & Chemical Technology and his Ph.D. in Chemical Kinetics & Reaction Modelling from the University of Calcutta. He had served the Government in several capacities starting as the Examiner of Patents & Designs, followed by Assistant Development Officer in the erstwhile DGTD, Deputy Director and Director in the Bureau of Industrial Costs & Prices, and Project Officer and Deputy Development Commissioner in the Ministry of Chemicals & Fertilisers. He had also worked for a couple of years in the Pharmaceutical Industry dealing with large-scale fermentation of antibiotics, production of sera & vaccines, and manufacture of various pharmaceutical formulations. During the last 13 years he has been working with the Ministry of Science & Technology, Department of Biotechnology, initially as Director, and for the last five years as an Advisor. His present assignment comprise development of biotech industry, issues related to biosafety of genetically modified organisms, patenting in biotechnology and providing extramural support to R&D Institutes involved in research on application-oriented biotech products.

Dr. Ghosh has made significant contributions in issues related to biosafety aspects of Genetically Modified

Organisms (GMOs) in India. He has been instrumental in updating the Recombinant DNA Biosafety Guidelines for the country. He played a key role in the initial phase of Industry-Institution interaction in Biotechnology, and a number of indigenous technologies had been transferred by him to the industry; some of these have been successfully absorbed and translated by the latter.

Dr Ghosh was inducted as a Member of the Bureau of Inter-Governmental Committee (ICCP) for the Cartagena Protocol on Biosafety in March 2000 to represent Asia. He also represented India in the Meetings of the Ad-hoc Working Group of the Conference of Parties (comprising nearly 150 countries) since 1996, for framing the Global Biosafety Protocol for the Trans-boundary Movement of Genetically Modified Organisms (GMOs) across different countries.

Dr. Ghosh has also significant interest in research and he has carried out R&D work on polymeric hydrophilic nanoparticles. He has a US Patent on polymeric hydrophilic nano-particle technology, which has been licensed to a reputed pharmaceutical company. Dr. Ghosh has published more than sixty scientific papers in various National and International Journals of repute. He has also authored two books. He is a Fellow of the Institution of Engineers (India) and a Fellow of the Royal Society of Chemistry, UK. He is also member and founder member of a number of learned societies. He is the recipient of Udyog Bandhu award from the Indian Chamber of Commerce and Industry.

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Introduction :

India has now more than 1000 million people, of whom nearly 340 million are very poor and are chronically under nourished. There has been increase in the *per capita* agricultural production over the years particularly preceded by a successful green revolution in seventies, but these successes have not been able to provide the minimum requirement of balanced quantities of food and nutrition to all our people. We need a collective will to achieve this. There can be no respite till the number of people below the poverty line is reduced to nil. In the context of India's increasing population growth and the limited possibilities of having enough additional land for agriculture, daunted by the country's lower technological levels, the questions that hover around are : Who are going to feed and clothe the large Indian population, particularly in the future years? How does India plan strategically to increase its food and fibre production? What are the options in hand? And what are the best choices for India?

There are many ways by which agricultural production can be raised. Modern technologies can and will be deployed

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* The views expressed in this paper are those of the author and they have nothing to do with the organisation to which the author belongs

to increase productivity. The current degradation rate of soil needs to be contained by use of bio-fertilisers; measures are to be taken for the conservation of top soil; strategies are to be adopted for the containment of bio-diversity; actions are needed for the introduction of improved measures for the control of pests and diseases; and efficient systems are to be introduced for the management and conservation of water. It is surmised that of all the measures, the application of modern biotechnology, integrated into the traditional system of agriculture in the rightful manner, may hold the key to augment our agricultural productivity on a sustainable basis.

In plants, absolute yield contributions[1] are met from the following factors:

	Factors	Contribution in %
1.	Genetic make-up of plants and optimisation of genes in them	50-60%
2.	Agronomic practices and agricultural technologies	25-30%
3.	Biotic and abiotic stress related factors	20-25%

It can be seen that maximum contribution is made by the genetic make of the plant cultivars.

There are presently three general approaches of applications of biotechnology as stated briefly below :

i) By selecting improved varieties through genome mapping to identify and propagate high yielding cultivars; developing somatic embryos of good varieties and micro-propagating them to generate true-to-types in large quantities; utilisation of anther/pollen culture to speed up propagation of high yielding new varieties etc.

ii) Harnessing near–full potential yields by developing cultivars resistant to viruses, bacteria, fungi and pests, tolerant to herbicides, salinity, drought, heat and water logging, etc.

iii) Improving existing products by directing production of economically more valuable products produced by other methods, e.g. converting rape seed/mustard to produce higher quantities of lauric acid otherwise obtained from coconut oil, converting soybean to produce more essential amino acids and reducing the content of enzymes responsible for interfering with trypsin metabolism, transforming sunflower to produce higher oleic acid, lengthening the period of maturing of the sorghum plant to increase its feed qualities, reducing the contents of anti-nutritional substances from tomato, delaying the ripening of fruits to improve their keeping qualities, modifying cotton cultivars to improve the fibre qualities such as better moisture absorption properties, etc.

The main advantages of utilising biotechnology in agriculture are the possibilities of increasing productivity through the use of newer varieties that possess such properties as resistance to pests, diseases and other stressful conditions such as drought, salinity or water logging. Of these measures, imparting the property of insect (specific) resistance through the transfer of a gene from *Bacillus thuringiensis* (Bt) into target plants by modern biotech methods is presently considered to be one of the most advanced biotechnology applications. Several such plants, commonly called as Bt-plants, are presently being commercialised in some parts of the world. To our knowledge, there are already more than 2000 Bt-strains that have been identified and about 50 genes coding for crystalline proteins that are toxic to certain insects and pests have been described. These crystalline proteins are commonly known as Cry proteins. Twenty-eight Cry protein-coding genes have been described in the literature out of which the genes coding for Cry1A, Cry1A (b), Cry1A(c), Cry1E and Cry3A (a) have been extensively used. The Bt-proteins selectively act on certain insect pests such as caterpillars (*Lepidoptera*), beetles (*Coleoptera*), flies & mosquitoes (*Diptera*). The genes effective against caterpillars and beetles are used to make Bt-plants.

There are about 70,000 pest species worldwide that damage agricultural crops. Of these, nearly 9,000 species are insects and mites. In this context, it is expected that the use of Bt-crops will increase productivity as well as provide significant benefits to the farmers, to the consumers, and to the environment.

It is estimated that chemical insecticides worth nearly US $ 8,800 million were spent globally for the containment of insects and pests during 1999 and that the share of such insecticides was about 30% for vegetables and fruits followed by cotton (22%), rice (16%), maize (8%) and other crops (24%). In India the pesticides consumption was worth about Rs 2800 crores in agriculture, and the major expenditure was in cotton.

Efforts are being made in India to harness the properties of Bt genes to reduce the application of chemical pesticides. If it happens, this phenomenon will also reduce the health risks of agricultural workers. Besides, the environment is also expected to be improved, as the harmful effects from the use of the broad- spectrum chemical pesticides will be reduced by the reduction in the use of chemical pesticides. India is experimenting with Bt-plants of cotton, tomato, cabbage, cauliflower, potato, tobacco and brinjal.[2] Good progress has been made on the contained open field experiments with these plants. Table 1 gives the status of research on these plants in India. It will be seen from the

Table that maximum progress has taken place in the case of Bt-cotton and herbicide resistant Indian mustard. In this paper the progress made in research on the Bt-cotton plants in India is specifically discussed.

Bt-Cotton Plants :

India plants nearly 9 million hectares of land under cotton and produces nearly 170 lakh bales (2.86 million tons) of cotton lint annually. The country is the third largest producer of cotton in the world. India currently contributes[3] to nearly 15.2% of the global production (Table 2). Of the seed cotton produced by the plant, nearly 35% is cotton lint and 65% is cottonseed. The average yield of cotton lint in India presently works out to about 320 kg per hectare. The national average has varied between 200-330 kg per hectare during the last 5 years. The average productivity from the hybrid seeds of cotton works out to about 600 kg while that from the non-hybrids is below 200 kg per hectare. Compared to the above, the Chinese average yield of lint is presently about 943 kg per hectare. There has, however, been steady rise in production and productivity in India over the years as can be seen[4] from the data in Table 3. Our neighbour country China has made better progress over the years as can be seen from the comparative yield data[3] presented in Table 4.

Cottons Cultivated in India :

In India, presently four cotton species are cultivated. These are known as *Gossypium hirsutum, Gossypium barbadense, Gossypium arboreum and Gossypium herbaceum*. The first two species are tetraploids and are inter-crossable while the next two species are diploids. The first two species (tetraploid) constitute 80% of the area under cotton production in India while the other two species constitute 20%. Hybrids are made by crossing two different lines of *Gossypium hirsutum*

Table 1
Indian developments in transgenic research and applications

Institute	Plants/ crops used for transformation	Transgenes inserted	Aim of the project and progress made
Central Tobacco Research Instt., Rajahmundri	Tobacco	Bt toxin gene Cry1A(b) and Cry1C	To generate plants resistant to *H.armigera* and *S.litura*. One round contained field trial completed. Further evaluation under progress.
Bose Institute, Calcutta	Rice	Bt toxin genes	To generate plants resistant to lepidopteran pests. Ready to undertake Green House testing.
Tamil Nadu Agricultural Univ., Coimbatore.	Rice	Reporter genes like *hph* or *gus A*	To study extent of transformation.
Delhi University, South Campus, New Delhi	Mustard / rape seed	*Bar, Barnase, Barstar*	Plant transformations completed and ready for green house experiments
	Rice	Selectable marker genes e.g. *hygromycin* resistance and *gus*	Gene regulation studies. Transformations completed.

Institute	Plants/ crops used for transformation	Transgenes inserted	Aim of the project and progress made
Indian Agricultural Research Institute sub-station at Shillong	Rice	Bt toxin gene	To impart lepidopteran resistance, transformations in progress.
Central Potato Research Institute, Shimla	Potato	Bt toxin gene	To generate plants resistant to lepidopteran pests. Ready to undertake Green House trials.
M/s Proagro PGS (India) Ltd., New Delhi	Brassica/ Mustard	*Barstar, Barnase, Bar*	To develop better hybrid cultivars suitable for local conditions; over 15 locations contained field trails completed. Further contained open-field research trials in progress at multi-locations.
	Tomato	*Cry*1A(b)	To develop plants resistant to lepidopteran pests; glass house experiments and one season contained field experiment completed. Further experiments in progress.
	Brinjal	*Cry*1A(b)	To develop plants resistant to lepidopteran pests; glass house experiments in progress

Institute	Plants/ crops used for transformation	Transgenes inserted	Aim of the project and progress made
	Cauliflower	*Barnase, Barstar* and *Bar*	To develop hybrid cultivars for local use; glass house experiments in progress.
	Cauliflower	*Cry*1H/*Cry* 9C	To develop resistance to pests; glass house experiments in progress.
	Cabbage	*Cry*1H/Cry9C	To develop resistance to pests; glass house experiments in progress
M/s MAHYCO, Mumbai	Cotton	*Cry1*A(c)	To develop resistance against lepidopteran pests; Multi-centric field trials in over 40 locations completed and further contained field trails in progress.
M/s Rallis India Ltd., Bangalore	Chilli	Snowdrop (*Galanthus nivalis*) Lectin gene	Resistance against lepidopteran, coleopteran & homopteran pests; transformation experiments in progress.

Institute	Plants/ crops used for transformation	Transgenes inserted	Aim of the project and progress made
	Bell pepper	Snowdrop (*Galanthus nivalis*) Lectin gene	Resistance against lepidopteran, coleopteran & homopteran pests; transformation experiments in progress.
	Tomato	Snowdrop (*Galanthus nivalis*) Lectin gene	Resistance against lepidopteran, coleopteran & homopteran pests; transformation experiments in progress.
Indian Agricultural Research Institute, New Delhi	Brinjal, Tomato, Cauliflower	Bt gene Bt gene Bt gene	To impart lepidopteran pest resistance, transformation completed, green house trials completed and one season field evaluation completed for Brinjal and Tomato.
	Mustard/ rapeseed	Arabidopsis annexin gene	Transformation completed, Green house trial completed, ready for field-trials for moisture stress resistance.
Jawaharlal Nehru University, New Delhi	Potato	Gene expressing for seed protein containing lysine obtained from seeds of *Amaranthus* plants(Ama-1 gene)	Transformation completed and transgenic potato under evaluation.

or by crossing lines of *Gossypium hirsutum* with *Gossypium barbadense*. The area under hybrids is about 50% of the total area of tetraploid cottons. Some of the main hybrids sold in the market are H.4, H.6, H.8, H.10, DCH.32, NHH.44, MECH.1, MECH.11, RCH.2, and ANKUR-615. These hybrids constitute about 50% of *Gossypium hirsutum* area, and the remaining 50% area of tetraploid cotton is covered by varieties such as LRA.5166, MCU.5, RAJAT, J.34, HS.6, B.N., F.846, F.505, F.1378, RST.9, LRK.516, etc. seeds of which are supplied by both private seed agencies and the State Seed Corporations.

Cotton is essentially grown as a Kharif crop in major parts of India. The land utilised for cotton cultivation can be divided into three distinct cultivation zones. These are the Central Zone comprising the States of Gujarat, Maharashtra and Madhya Pradesh, the North Zone comprising Punjab, Haryana and Rajasthan, and the Southern Zone comprising Karnataka, Tamil Nadu and Andhra Pradesh. The Central Zone contributes to nearly 55% of the total land use for cotton while each of North and Southern Zone contributes to 22-23%. The Northern Cotton Zone is predominantly irrigated, and in terms of usage of cotton cultivars it is almost homogenous and it utilises high yielding varieties for planting. Presently, hybrids are also being utilised in parts of Northern Zone. In the other two zones, the cultivation patterns are quite heterogeneous. In major parts of the country like Punjab, Haryana, Rajasthan, Uttar Pradesh, Gujarat, Madhya Pradesh, Maharashtra and parts of Karnataka as well as Andhra Pradesh, the irrigated crop is sown in March- May while the rainfed crop is sown in June-July. In parts of Gujarat and Uttar Pradesh, pre-monsoon dry sowing is also practised towards the end of May or early June. In Tamil Nadu, the planting is carried out in September/October, but in Southern districts the sowing extends upto November. In parts of Andhra Pradesh and Karnataka, desi cotton is

Table 2

Country-wise production, productivity and market share of cotton : 1998-99

Country	Production (million tonnes)	Area (million hectares)	Productivity (kg/hectare)	Market share in %
China	4.300	4.56	943	24.5
USA	4.132	5.37	769	16.5
India	2.856	8.90	321	15.2
Pakistan	1.593	2.89	552	7.5
Turkey	0.755	0.71	1065	4.6
Egypt	0.315	0.36	873	1.3
World	**19.735**	**33.82**	**584**	**100**

Table 3

Cotton in India : Year-wise land use, production, yield and percent land coverage under irrigation

Year	Area (million hectares)	Production (million tonnes)	Yield (kg/ha)	% land coverage under irrigation
1951-52	5.89	0.530	88	9.1
1961-62	7.98	0.780	103	13.0
1971-72	7.80	1.119	151	20.3
1981-82	8.06	1.428	177	27.7
1991-92	7.66	2.023	264	33.3
1996-97	9.12	3.004	330	35.0
1998-99	9.30	2.780	298	37.0

sown in August and September. In terms of contribution in production, usually Maharashtra and Gujarat combined top the production; this is followed by Andhra Pradesh, Haryana, Punjab and Rajasthan.

Main Market Player of Cotton Seeds :

Presently India uses nearly 25000 tones of cottonseeds annually for planting, out of which 4500 tones are hybrids and the rest are certified varieties and own seeds. Among the main market players of hybrid and varietal cottonseeds, Rasi, Ankur, Mahyco, Mahindra, Ajeet Seeds, Vikram Seeds, and Nuziveedu Seeds and State Seeds Corporations of Maharashtra, Gujarat, Andhra Pradesh and Karnataka are the most important ones.

Chemical Pesticides Usage in Cotton :

India uses annually nearly Rs. 3200 crores worth of

Table 4
Comparative production and yield of cotton : India Vs China

Year	India		China	
	Production (million tonnes)	Yield (kg/ha)	Production (million tonnes)	Yield (kg/ha)
1951-52	0.530	88	1.30	240
1961-62	0.780	103	0.75	225
1971-72	1.119	151	2.33	420
1981-82	1.428	177	2.70	555
1991-92	2.023	264	4.50	660
1996-97	3.004	330	4.30	943
1998-99	2.780	298	4.30	943

chemicals which include pesticides for the control of mosquitoes and sand flies besides agricultural pesticides and herbicides. Out of Rs. 2800 crores worth of chemical pesticides used in agriculture, Rs. 1600 crores worth are consumed on the cotton crop alone for the control of bollworms and sucking pests. It is estimated that about Rs. 1100 crores worth of chemical pesticides are used only for the control of bollworms in cotton. Government wants to examine all possible ways of reducing pests-linked damage in cotton crop, which can result into substantial savings for the cotton farmers. This is precisely the reason for India's experimenting with Bt-cotton. It is also planned to examine if transgenic Bt-cotton is environmentally safe, and if this would contribute to increase in yield and at the same time would contribute to substantial reduction in chemical pesticides consumption.

About Bt-Cotton :

Bt-cotton, presently under field experimentation is the genetically manipulated cotton that produces a protein, which is toxic to lepidopteran insects when ingested in adequate quantities. From the limited results obtained in the field, it has been observed that Bt-cotton provides excellent control of the key caterpillar pests in Indian cotton fields, such as the American Bollworm (*Helicoverpa armigera*), the Spotted Bollworm (*Earias vittella*), the Spiny Bollworm (*Earias insulana*) and Pink Bollworm (*Pectinophora gossypiella*).

The toxin in Bt-cotton exists in nature within the microorganism *Bacillus thuringiensis*. It was first discovered by the Japanese bacteriologist Ishiwata Shigetane in 1901. Subsequently, in 1915, a German scientist, named Ernst Berliner, isolated this toxin from a dead moth in Thuringen region of Germany. The bacterium has thus been named as *Bacillus thuringiensis (Bt)*.

The genetic manipulation of cotton plants has been carried out by the insertion of a gene known as Cry1Ac gene obtained from the bacterium *Bacillus thuringiensis*. The natural gene has been partly modified to improve its properties. The plant releases a protein, which is a special type of Bt-protein that is very specific in toxicity against the lepidopteran pests. These insect larvae eat the various parts of the transgenic plant and die. However, Bt-cotton is not effective against non-target pests such as sucking pests, beneficial insects and predators. Therefore, additional pesticides may have to be applied for effective pest control when pest pressure is very high or if such insects are present which are not controlled by the Bt-cotton. Table 5 indicates the various insect pests of cotton found in India.[5,6]

Use of Bt-cotton is expected to reduce the use of chemical insecticides in the cotton crop. In addition, as the protein acts only on target pests (Lepidopteran pests), it will not reduce the population of beneficial and predator insects, which also help to keep the destructive pests population in control.

Bt-cotton is also seen to have some control against tobacco caterpillars (*Spodoptera litura*), but it does not kill sucking pests such as the Thrips, Jassids, Aphids, Whitefly etc., beneficial insects and natural predators like Honeybees, Green Lacewings, and Ladybird Beetles.

The modified gene Cry 1Ac was developed by Monsanto Inc., USA and the transgenic cotton varieties containing this gene have been designated as Bollgard™ cotton by the company. Other Bt-cotton varieties are also under development in different parts of the world. China has its own Bt-cotton, which contains a Cry1Ac gene that is different from that of Monsanto. India is also trying to develop its own Bt-cotton with a new Bt-gene. Presently the Bt-cotton is commercially used in the USA, Australia, China, Mexico, Argentina and South Africa and is under development in Brazil, Zimbabwe, Turkey, as well as in India, as stated above.

The total land used for cotton cultivation in the world in 1999 was about 33.8 million hectares. The main countries in cultivation included India, China, USA, Pakistan, Brazil, Argentina, Tanzania, Greece, Egypt, Paraguay, Nigeria, and Zimbabwe. The maximum use of Bt-cotton in 1999 was seen in the United States of America where more than 55% of the total cotton area of 5.9 million hectares was covered under transgenic cotton, of which Bt-cotton occupied 1.7 million hectares.

Bt-cotton provides no control during the egg laying stage of lepidopteran pests. The larval stage of the insect is the most important stage, as damage occurs to the plant during this period. After emerging from the eggs, the larvae feed on the plant to grow in size. If they are not controlled when they are small, they will eventually damage the cotton bolls, causing them to rot. When the larvae feeds on Bt-cotton, it ingests a lethal dose of the Bt-protein and the larvae die within about three days. Bt-cotton is found to provide exceptional control of target pests at this stage.

It is important to clearly identify the types of larvae on the plant, as Bt-cotton does not control all insects. The larvae are best scouted for on both sides of leaves, inside the stems, and inside flowers, squares, and bolls.

Scouting for insect pests is not only necessary for destructive pests, but it is also necessary for beneficial insects. Beneficial insects, predators and parasites like Hymenoptera, Honeybees, Green Lacewing and Ladybird Beetles are not either harmful to the plant or they destroy harmful pests. If the level of beneficial insect population is high, then it can be expected that the number of destructive pests be reduced

Table 5
Major insect-pests of cotton prevalent in India

Common Name	Plant-infestation period	Regional-distribution	Remarks
Bollworms			
American bollworm	Vegetable phase- Flowering phase	All Zones	Key pest
Pink bollworm	Flowering phase	North Zone South Zone	Minor pest Key pest
Spotted bollworm	Vegetable phase Flowering phase	All Zones	Key pest
Sucking Pests			
Thrips	Cotyledonary-phase	North Zone South Zone	Minor pest Key pest
	Flowering-phase	Central Zone	Minor pest
Leafhopper	Vegetable-phase Flowering-phase	North Zone Central Zone South Zone	Key pest Key pest Minor pest
Whitefly	Vegetative phase Boll bursting phase	All Zones	Key pest
Cotton aphid	Vegetative phase Boll bursting phase	North Zone	Minor pest
Foliage Feeding Pests			
Cotton leaf roller	Flowering phase	North Zone	Sporadic pest
Green semi looper	Flowering phase	North Zone	Sporadic pest
Black semi looper	Vegetative phase	North Zone	Sporadic pest
Bihar Hairy caterpillar	Flowering phase	North Zone	Sporadic pest
Tobacco caterpillar	Flowering phase	North Zone South Zone	New pest Key pest
Leaf Surface weevil	Vegetative phase Flowering phase	Central Zone	Minor pest
Leaf miner	Seedling phase Vegetative phase	All Zones	News pest
Leaf perforator	Vegetative phase Flowering phase	South Zone (Tamil Nadu)	Minor pest

Common Name	Plant-infestation period	Regional-distribution	Remarks
Soil Pests			
Termite	Cotyledonary phase Flowering phase	North Zone	Minor pest
Cutworms	Cotylednary phase	North Zone	New pest
Miscellaneous Pests			
Shoot-Weevil	Vegetative phase Flowering phase	South Zone (Karnataka)	Minor pest
Stem-Weevil	Vegetative phase Flowering phase	South Zone (Tamil Nadu)	Key pest

significantly. In such cases, spraying should be done only if the beneficial insect population cannot effectively control the destructive insect population. Surveillance through scouting therefore becomes very essential. Premature spraying often kills the beneficial insect population that is effectively controlling the destructive pests.

While Bt-cotton provides very effective control when target pest populations are normal or low, if pest infestations are high, one to two supplemental sprays for target pests may be needed which should be based on economic threshold levels. Much work has been done to determine the Economic Threshold Levels (ETL) in cotton against different insect pests in India. Table 6 gives the ETL values for different cotton pests in India.[5,6]

Cotton as a Perennial Crop :

Cotton is a perennial crop, which can continues to yield cotton after the first season. Most farmers cease to harvest bolls after one season as the cotton plant ages, because the productivity of the plant falls dramatically. In most of the circumstances, it makes more sense to plant another crop

Table 6

Economic Threshold Level (ETL) Values of major insect pests of cotton in India

Insec1t-Pests	Central Zone	North Zone	South Zone
Bollworm complex	5% incidence level in retained and shed fruit bodies	5% incidence in shed fruiting bodies	10% incidence level in retained and shed fruiting bodies
Pink bollworm		5%affected fruiting bodies 5% affected green bolls 4-8 moths/trap/ night	
Spotted bollworm		5% affected twigs/ shoots 5% incidence in shed bodies	
American bollworm		5% incidence in shed bodies 0.5 larvae/plant	7 moths/trap/ night
Thrips		10-thrips/leaf 20-30%infested plants	10-thrips per leaf, 2 nymphs or adults/ leaf
Jassid	2 nymphs per leaf	2 nymphs per leaf Appearance of second injury grade on 50% plants	2 nymphs per leaf
Whitefly	8-10 adults per leaf	6 adults per leaf 20 nymphs/leaf Appearance of honeydew on 50% plants	8-10 adults or 20 nymphs per leaf
Aphids	15-20% affected plants	10% infested plants Appearance of honey dew on 50% plants	15-20% affected plants

rather than to continue as a ratoon crop, as the yield of cotton comes down substantially in ratoon cropping. We observed significant reduction in the capacity of certain Bt-cultivars to resist the attack of lepidopteran pests on ageing of such Bt-cotton plants.

Regulatory Structure to Conduct Field Trials using Genetically Modified Cotton Plants :

All experiments on genetically modified cotton (GM-cotton) are controlled under the Indian Environment (Protection) Act, 1986 (EPA). The "Hazardous Microorganisms Rules" for handling of GM-crops were announced in 1989. The working arrangement is that all organisations in the country dealing with GM-crops should obtain permission from its "Institutional Bio-Safety Committee" (IBSC). All IBSCs have a DBT-representative to oversee if all the safety procedures are being followed. Only less risky experiments can be authorised by IBSC. But for risky experiments such as experiments with Bt-cotton, IBSC sends the proposals to the central committee know as the "Review Committee on Genetic Manipulation" (RCGM). The RCGM grants permits for field experiments in small plots. Experiments are to be conducted with utmost care under contained conditions. The RCGM had prepared the latest guidelines in August 1998, which was subsequently revised further to incorporate amendment upto September 1999. RCGM has constituted a "Monitoring-cum-Evaluation Committee" (MEC) with eminent agricultural scientists as its members. MEC conducts visits of the experimental field to evaluate if the experiments are conducted according to the Rules and Procedures, and if the crops in the fields have agronomic advantage. The RCGM also authorises generation of data on "Food Safety" in labs using adult laboratory animals. After the small field experiments are completed and the food safety data are generated to the satisfaction of RCGM, the information is collated in the form of a document called as the "Registration Document". This document is either called for by the RCGM or is placed by the applicant before the "Genetic Engineering Approval Committee" (GEAC) in the Ministry of Environment and Forests. GEAC is responsible for the approval of activities involving large-scale use of hazardous and genetically engineered organisms including GM-crops for further large-scale research or for commercial production. The experiments are conducted scientifically and these are monitored very meticulously.

Field Experimentation of Bt-Cotton in India under EPA :

The purpose of field experimentation is to assess Bt-cotton from environmental safety issues. Field experimentation is not equivalent to commercial use. All care is taken to ensure that during field experimentation the Recombinant DNA safety guidelines of the government are followed and experiments are so designed as to minimise the risks to the environment, the land and the people exposed to the experiments.

An Indian company namely M/s. Maharashtra Hybrid Seeds Co. Ltd., (Mahyco), Mumbai, in which presently a foreign company M/s. Monsanto Enterprises Pvt. Ltd. Mumbai, (which is a 100% subsidiary of Monsanto Inc., USA) has 26% equity, was permitted by the government to undertake experiments on Bt-cotton in the country.

The Government has permitted field experimentation of Bt-cotton varieties to Mahyco to understand how these varieties behave in different agroclimatic regions.

Initially on March 10, 1995, the Department of Biotechnology (DBT) had permitted the import of 100 g of transgenic Cocker-312 (a variety of cottonseed cultivated in USA) to Mahyco. The seeds contained the Bt Cry1Ac gene. The applicant company imported the seed and back crossed the trait into elite Indian varieties for six generations and saved seeds in each generation for next experiments in the contained green house. After at least four back crossings, the subsequent generations were selfed to generate stable lines to be used for making hybrids. The experiments conducted in the field were with hybrids derived from lines generated after four back crosses and two selfed generations. The Indian hybrids designated as MECH.1, 3,12,15, 160,162,184 and 915 have been bred to contain the Cry1Ac gene, and these genetically modified cotton hybrids have only been permitted for field evaluation along with non-Bt hybrids, national checks and local checks as the controls.

Limited field trials were conducted in Andhra Pradesh, Karnataka, Maharashtra, Tamil Nadu and Haryana, in November 1997, in plots of 20 m × 10 m at each location. Subsequently, slightly larger areas at 40 locations were allocated in the states of Andhra Pradesh, Maharashtra, Gujarat, Karnataka, Haryana, Punjab, Tamil Nadu, Madhya Pradesh and Rajasthan during August 1998 for conducting these trials. In 1999, eleven additional field trials were conducted. The MEC constituted by the RCGM had conducted visits of the experimental sites. The State Governments have been kept informed each time the experiment-permits were issued.

The results of environmental safety studies including gene flow studies of the pollens from the transgenic Bt-cotton, the aggressiveness of the Bt-plants, their crossability, effect of the pollens and plants on non-target organisms and the studies of the exposure of the transgenic proteins did not indicate any substantial difference of the transgenic plants from the non-transgenic ones. The gene flow studies indicated out-crossing of non-transgenic plants from 0.42% to 2.1% up to a distance of 2 metres only; these results are consistent with earlier studies conducted in India and USA.

The Bt-plants were ratooned and irrigated at certain locations at regular intervals to allow germination of any spilled over seed after boll bursting. After set intervals, the germinated plants were counted to assess if there was any difference between their numbers in the Bt-cotton field compared to the controls in the non-Bt cotton field. There was no significant difference between the transgenic and non-transgenic cotton with regard to aggressiveness. Cotton pollens were not found to be compatible for cross-pollination with any near relatives other than cotton in Indian environment. The tetraploid Bt-cotton pollens had travelled to short distances to out-cross the non-transgenic tetraploid controls as was observed in three seasons of studies at five locations across the country. Consequently, it was considered that the possibility of out-crossing other genetic resources by the transgenic Bt-cotton pollen was remote. The Cry1Ac proteins produced in Bt-cotton is highly toxic to targeted insect pests. In a study elsewhere, the Bt-cotton plants were evaluated against two soil beneficial insect species of the order *Collembola*. The experiments demonstrated that the Bt-proteins have no detrimental effects on the survival and the production of *Collembola* species. In another study conducted in Greece with Bt-cotton plants, it was observed that the presence of *Collembola* and earthworms population did not change in the Bt-cotton plots compared to the controlled plots.

It can therefore be stated from these observations that there is no different risks from the use of Bt-cotton plant to the environment than from the use of the non-Bt-cotton plant.

The experimental Bt-cotton plots required less number of sprays of chemical pesticides. Indeed, in some plots no spraying was required while in some upto two sprays were adequate. In non-Bt plots as many as nine to twelve sprays were required for effective control of the pests.

Food Safety Evaluation of Bt-cotton :

Food safety evaluation of seeds, oil, and cake of Bt-cotton has been done simultaneously to establish their safety to humans and animals. The studies conducted at the Industrial Toxicology Research Centre (ITRC), Lucknow have shown that Bt-cotton is safe to ruminants. The earlier available data from the collaborators in USA had shown that Bt-cotton is safe to different animals including mammals, birds and fish. The data on allergenicity studies conducted on Brown Norway rats have shown that there is no allergenicity developed in these animals by the use of Bt-cotton.

The results of the field trials and safety studies over the last four years have helped the Indian government to assess the economic benefits and safety of Bt-cotton and would enable the government to take suitable steps for moving to the next phase such as for large scale trials and seed production.

The Controversy of Terminator Gene in Bt-Cotton :

There is no "Terminator Gene" in the Bt-cotton, which is under field-testing. As stated earlier; the transgenic seeds of cotton which are in the field have been developed after at least four backcrosses and two selfing generations, which would imply that these seeds have progressed up to at least six generation in India. If the seeds contained the so-called "Terminator gene", any next generation after crossing would not have produced the viable seeds. There is no scientific evidence in the news that appeared in Indian press and media that the Bt-cotton seeds used in Indian experiments contained "Terminator gene". The Indian Government has already banned the import and use of seeds containing the "Terminator gene".

Concluding Remarks :

Genetically modified plants are expected to contribute to increase in production in Indian agriculture. Presently limited field experiments are being performed with Bt plants of cotton, tomato, cabbage, cauliflower, potato, tobacco and brinjal. Based on the progress of experiments made in the Indian soil on Bt-cotton, it appears that it has great potential in increasing the yield of cotton, and in reducing the consumption of chemical pesticides. The product also seems to pose no severe problem to the environment and to the food and feed safety issues of human and animals. Most of the environmental safety issues have been resolved through the present scientific knowledge base. Some more experiments are presently being conducted.

The world-wide use of Bt-cotton in commercial agriculture in 6 countries in 1999 exceeded an area of 1.9 million hectares. This fact also reinstates our belief that Bt-cotton might be found to be safe in our environment too.

It is believed that perhaps the greatest challenge to the acceptance of Bt-cotton by the society would not be with the plants themselves but by the high expectations from the farmers. As the science in its developments is well documented and is on strong foundation, the challenges are expected to be emanating from the high expectations of the farmers and the society. This may be fuelled by over promotion of the new product. Rumours may fuel expectations that Bt-cotton would control all insects without the use of any insecticidal sprays. It would be very important to educate the farmers and the dealers in the right way so that they could aim for realistic expectations from Bt-cotton. Farmers must be told that Bt-cotton does not control all pests. It reduces the target pests substantially, but it does not eliminate and eradicate all the pests. Consequently, there may be the need for the application of insecticides. To obtain the most effective control of pests while planting Bt-cotton, farmers must undertake extensive scouting of the field to decide and understand when supplemental sprays of chemical insecticides for control of cotton pests are necessary.

Some Indian farmers may choose to continue to grow cotton like a ratoon crop. As Bt-cotton ages, the level of Bt-protein expressed may fall. The gene expression may fall below acceptable control limits. Until complete field experimentation is conducted and conclusions drawn in favour, it would be proper to avoid perennial harvesting of Bt-cotton. Large scale field experiments must be carried out to find out the truth of economic benefits from ratoon harvesting.

From the field experiments conducted in India, it was found that there was an increase in the productivity ranging from 23-60% in 1998 experiments and 29-88 % in 1999. The controls were the farmers' practices. The initial results are thus encouraging.

Bt-cotton is in use in the manufacture of cloth fabrics for human for the last 4 to 5 years. Several countries are utilising such cotton for making fabrics and clothes. There has not been any reported allergenicity from the use of such clothes from anywhere in the world. Very low level (0.17 ppm) of Bt-protein was detected in one of the reports in raw linters.[7] Raw linters are used mainly in the processed form for industrial purposes. The processing reduces and degrades the Bt-proteins further below detection levels. The level of Bt-proteins present in raw linters is much lower than that found in Bt-plant parts like roots, stem, leaf, bolls etc.

From the Indian experiments carried out so far and from the published information available from elsewhere it has been found that the Bt-cotton plant is nearly equivalent to non-Bt-cotton plant in every respect, except that Bt-cotton plant has an additional property of producing its own biopesticide to protect it from its main target insect pests.

Bt-cotton plants yielded more cotton per hectare of land and required less chemical pesticides. About 60 million people are dependent on cotton cultivation, trade and processing in India. The export earnings from cotton textiles, yarns and garments amounted to over Rs. 30,000 crores in 1998-99, which holds the potential of growing further. The Bt-cotton technology may prove a boon for the economic upliftment of cotton farmers in the country. It is hoped that when in future such cotton plants are introduced, they would lead to reduction in chemical pesticides usage and would contribute to productivity increase that may lead to a cotton revolution in India.

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