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on

"Future Technologies : Indian Cotton in the Next Decade"

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BOOK OF PAPERS

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PREFACE

Cotton is one of the most ancient and very important commercial crop of global importance with a significant role in Indian agriculture, industrial development, employment generation and improving the national economy. It is cultivated for domestic consumption and also exported worldwide in about 111 countries worldwide and hence called "King of fibres" or "White gold". Millions of people depend on cotton cultivation, trade, transportation, ginning and processing for their livelihood. India is the only country in the world growing all the four cultivated species of cotton alongwith their hybrid combinations in the vast diverted agro-climatic situations. Cotton is basically cultivated for its fibre which is used as textile raw material. It is cultivated from Punjab in the north to Kanyakumari in the south and Assam in the east to Kutch (Gujarat) in the west.

India, the second largest producer, consumer as well as exporters of cotton next to China with 34 per cent of world area and 21 per cent of world production and continue to maintain the largest area under cotton. Within a span of thirteen years, the cotton production in the country has gone more than double with the increase of the productivity. The productivity of cotton has not made headway because of more than 70 per cent area is under rainfed cultivation and appearance of new diseases and insect-pests in transgenic cotton. However, new emerging threats in terms of biotic and abiotic factors are to be understood properly and effective strategies need to be evolved for their proper redressal. The problems and prospects of Bt cottons in the country need to be put in a proper perspective. Therefore, there is an urgent need to properly understand the IPR issues in the best interest of farmers and scientists.

In order to maintain pace with the increased demand for the commodity, both in national and international market, it is imperative to give impetus for development of new cotton varieties and hybrids with appropriate cultivation technologies. Introduction of large number of private sector Bt cotton hybrids have brought a welcome change in recent times as far as production gains are concerned. However, to meet the ever increasing demand both in the domestic and international markets, an effective strategy needs to be developed.

With continued advances in plant breeding, plant genome and biotechnology research, improved seeds have to be evolved for reaping the untapped yield potential. Good quality seed acts as a catalyst for realizing the potential of all other agriculture inputs. Continuous efforts are requires to make use of new technologies for efficient crop improvement and management.

The research papers included in the **"Book of Papers"** are related to **"Crop Improvement, Biotechnology and Post Harvest Technology", "Crop Production, Mechanization and Economic Development" and "Crop Protection and Biosafety"** which were the theme areas of the symposium. Present compilation on **"Future Technologies : Indian Cotton in the Next Decade"** is a compendium of holistic advancements and other relevant information related to cotton covering different disciplines. We hope that the information contained in this "Book of Papers" will be useful to all the stakeholders *viz.,* researchers, students, developmental officers, planners and farmers. All these manuscripts have been pre reviewed by eminent scientists of the for respective disciplines/fields before publishing in this "Book of Papers". We are thankful to the authors of individual chapters/papers for their contribution, time and diligence without which this volume would not have been possible.

We deem it a rare privilege to place on record our sincere gratitude to Dr. D. P. Biradar, President, CRDA for his valuable guidance and directions in the general functioning of CRDA. We take this opportunity to thank all concerned and hope this book of papers will serves the purpose of cotton research workers for furthering the cause of cotton farmers.

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Dated: 17-12-2015

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Book of Papers on "Future Technologies : Indian Cotton in the Next Decade"

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LEAD PAPERS

Diploid cotton improvement through interspecific hybridization- An account of pre breeding efforts

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Cottons are not only a world's leading textile fiber and oilseed crop, but also a crop that is of significance for foil energy and bioenergy production. Out of the 50 *Gossypium* species, four, including two *G. hirsutum* and *G. barbadense* allotetraploids and two *G. herbaceum* and *G.arboreum* diploids are cultivated. Upland cotton (*G. hirsutum),* known for, long staple cotton, orMexican cotton, produces over 90 per cent of theworld's cotton; *G. barbadense*, (Sea Island, American Pima, or Egyptian cotton) also known for extra long staple (ELS) cotton, contributes 8 per cent of the world's cotton; and *G. herbaceum*, also known as Levant cotton, and *G. arboreum*, also known as Tree cotton, together provide 2 percent of the world's cotton (Zhang, *et al.,*2008).

Upland cottons (*G.hirsutum* L.) account for over 90 per cent of lint production because of their high productivity and it is cornerstone of the textile industries worldwide. In addition, in India Asiatic cottons (*G. arboreum* and *G. herbaceum*) are known as *desi* were grown on about 98 per cent area around 1947 and the *G. hirsutum* on just around 2.Presently, the situation is now exactly the reverse. The diploids and their hybrids are cultivated on very less (0.50 lakh ha) area in which *desi* cotton hybrid contributes only 1 per cent in production. However, now a day even short and coarse staple of diploid cotton is in great demand, particularly in fabrics like denim and upholstery, filling and medical purpose as well as fetches attractive price also. So, why not promote *desi* cottons, which are resistant to drought, water logging, diseases and pests especially cotton leaf curl disease, a dread disease of *G. hirsutum*, well adapted to the climatic aberrations, suitable underrainfed conditions, wider adaptability and low cost of management? Thus *desi* cotton is to the rescue of Indian cotton growers (Mehetre,2015).

Among the diploid species (2n=2 x =26), *G. arboreum* and G. *herbaceum* are generally cultivated on marginal and drought prone regions of Asia. Phenotypically, they can be distinguished based on plant habit as well as leaf, bracteole and boll features (Fryxell, 1979). Long and narrow lobed leaves, bracteoles with fewer teeth and round tapering bolls are the characteristics of *G. arboreum*, while constricted leaf lobes, wide bracteoles and round, and less pitted bolls are the common features of *G. herbaceum*. Within the A genome*, G. herbaceum* and *G. arboreum* diverged relatively recently. Cytologically these species can be distinguished by a reciprocal translocation (Gerstel, 1953), while the At (A subgenome in tetraploid) differs from the A, D and Dt (D subgenome in tetraploid) genomes by two reciprocal translocations. This suggests that *G. arboreum* arose as an incipient species with the origin through the fixation of the translocation (Endrizzi *et al.,* 1985). Recently, draft sequences of the putative D genome parent G. raimondii (Wang *et al.,* 2012) and for

developmental traits, yield and fiber characters in *G. arboreum* (Singh and Singh, 1984) and G. *herbaceum* (Singh, 1983). Old world Asiatic diploid cottons were economically important during

A genome parent *G. arboreum* (Li *et al.,* 2014) have been sequenced, providing new insights into the divergence among the polyploidy species. Potentially valuable genetic variability has been observed early global expansion of commercial cotton production. In the 1950's with the introduction of new world cotton, the area under diploid cotton cultivation was drastically reduced. Diploid cotton, however, is a model system for studying the genetics of fiber development compared to the more complicated system in tetraploid cottons. Therefore an understanding of the genetic inheritance and genomic regions controlling the fiber genes of diploid cotton species is critical (Li *et al.,* 2014). In order to use the extant genetic diversity in the development of superior genotypes or transferring elite genes into cultivated tetraploids, molecular breeding techniques offer promising avenues compared to traditional breeding methods. Molecular linkage maps provide essential tools for plant genetic research, facilitating quantitative trait locus (QTL) identification, marker-assisted selection, and map based cloning. To date, several genetic maps of cotton genomes have been constructed using diverse molecular markers and different mapping populations in tetraploid cottons (Reinisch *et al.,* 1994 Ulloa *et al.,* 2002; Rong *et al.,* 2004; Mei *et al.,* 2004; Nguyen *et al.,* 2004; Han *et al.,* 2004 and Zhang *et al.,* 2009). Comparatively few genetic maps have been developed in segregating populations involving diploid species. Interspecific linkage maps of diploid cottons have been constructed for the A genome (G. *herbaceum* × G. *arboreum*), the D genome (G. *trilobum* × G. *raimondii*) (Brubaker *et al.,* 1999; Rong *et al.,* 2004; Desai *et al.,* 2006) and the G genome (G. nelsonii × G. australe) (Brubaker and Brown, 2003). An RFLP linkage map using an interspecific A genome diploid F2 population mapped 275 loci (Desai *et al.,* 2006). The 13 chromosomes of the A genome were represented by 12 large linkage groups reflecting an expected inter-chromosomal translocation between the parents. Though the diploid mapping parents are the closest living relatives of the allotetraploid At genome progenitor, two translocations and seven inversions were found between the A and At genomes (Desai *et al.,* 2006). Research on 'A' genome cotton has declined with the decrease in their cultivation during the last few decades. Although molecular tools have been available for a long time, only limited research has been carried out on the Asiatic cotton species (Brubaker *et al.,* 1999). Understanding the molecular genetics of A genome cotton can be important for many reasons. They can foremost serve as a simple model system to study complex quantitative traits, yet only a limited number of genetic maps and QTL studies have been conducted. There is a significant opportunity for further mining the diploid genome with efficient marker systems to facilitate genetic mapping of fiber genes. In the present study, we used AFLP, SSR and TRAP markers to generate a framework genetic map of cultivated diploid cottons. The genetic map was used to identify QTL linked to fiber traits.

Cotton is infected by several insects, pests and pathogens inducing different diseases. Among them cotton leaf curl virus (CLCuV) is the most damaging disease, causing enormous losses (Khan and Ahmad, 2005). With the passage of time, CLCuV has spread to all provinces of the Pakistan (Tariq, 2005). It has caused a reduction of 9.45 million cotton bales during the last decade. Reduction in yield of

tolerant and susceptible varieties is reported to be 50 and 85-90 per cent, respectively (Hussain, 1995; Khan *et al.,* 2001). A new recombinant strain of egomovirus derived from cotton leaf curl Multan virus (CLCuMV) and cotton leaf curl Kokhran virus (CLCuKV) has been found to be associated with the breakage of resistance in existing cotton varieties .

Asiatic (*G. arboreum* L.) varieties of cotton have built-in desirable genes for drought tolerance and resistance to insect pests such as leaf hoppers and diseases like black arm, root rot and reddening of leaves, hence they are suitable for dry land conditions and low input technology. On the contrary, American (*G. hirsutum*) varieties are high yielding and widely grown. However, they lack important characteristics prevalent in diploid cultivated varieties of cotton and their cost of production is therefore relatively high. It is worthwhile to combine the genes for the above-referred desirable characters between *G.hirsutum* and *G. arboreum* cotton. However conventional breeding methods, the crosses between these two species are unsuccessful due to abortion of embryo after fertilization because of differences in chromosome numbers of *G. arboreum* and G. herbaceum (2n= 26) and *G. hirsutum* (2n=52) cotton. Hence embryo culture method to overcome such situation in production of interspecific hybrids was attempted. Number of incompatibility barriers is major bottleneck in introgression of genes from wild/cultivated diploid to cultivated American cotton. Attempts were made to obtain the interspecific hybrid between cultivated *G. hirsutum* and *G. arboreum* through *in vitro ovule* culture. Very limited success has been reported by this advent technique. Even if crossing is successful and hybrids, if obtained, the resultant progeny will be triploid with sterility. Only after doubling of chromosome complement, it can be backcrossed to cultivated tetraploids to obtain plants with 2n = 52 chromosomes having desired character combinations. Direct crossing of induced tetraploids of the *G. arboreum* L. and *G. herbaceum* L cottons with cultivated tetraploids *G. hirsutum* and *G. barbadense* would be a shortcut method for transfer of desirable genes from cultivated diploids to tetraploids and vice versa. Considering limitations in interspecific transfer of *G. hirsutum* characters like bigger boll size and superior fibre qualities in present study attempts were made to induced autotetraploid of *G. arboreum* and *G. herbaceum* with 2n=52 chromosomes and then cross with them with *G. hirsutum* cotton. Reports on successful induction of polyploidy in *Gossypium* by colchicine treatment are reported earlier. Hybridization between autotetraploid *G. arboreum* (2n=4x=52) and *G. hirsutum* (2n=4x=52) was reported successful (Deshpande *et.al.,* 1991 and Mehetre *et. al.,* 2003,c).

However, the negative correlation between lint yield and fibre strength (Guo, *et. al.,* 2003) limits the success of selection programme to improve both yield and a fibre properties. Genetic association of these traits is caused by either linkage or pleiotropism (Narayanan,1972) due to which, even after three cycles of recurrent selection though the lint yield was increased, the fiber strength was not proportionately than that of the base population. Random intercrossing suggested (Narayanan, 1977) to reduce negative genetic correlations if exist because of linkage. With random intercrossing of populations derived from the tri-species hybrid (*G. arboreum* L. x *G. thurberi* Tod. x *G. hirsutum, L.*), resulted in a reduction of correlation between yield and fiber strength (Narayanan, and Sreerangasamy, 1973 and Memon and Ahmad, 1970). Genotype

environment interaction is larger for lint yield than for fiber strength (Meyer, 1972, 1973).Further, the lint yield and fiber strength is conditioned by additive and non-additive gene action whereas fiber strength is mostly additive gene action (Fryxell,1976), fiber strength far high heritability while yield has low heritability. Thus improved desirable genetic combinations of yield and fibre strength genes are achieved by adapting backcrossing (Narayanan, 1972) .

Production/isolation of haploids in *Gossypium*, L. species : The utilization of haploids (2n=2x=26) of tetraploid (2n=4x=52) cultivated cotton species in interspecific hybridization with wild diploids offers scope for minimizing the period for interspecific transfers. Potential of haploids in interspecific cotton breeding and in basic and applied research is discussed. Efforts were made at Mahatma Phule Krishi Vidyapeeth, Rahuri to induce and isolate haploids from experimental and natural as well as segregating populations of populations **interspecific (*G hirsutum* x *G. barbadense*), respectively and utilized them in interspecific hybridization with diploid wild and cultivated species of *Gossypium*.**

The details of the haploids obtained and studied are as under

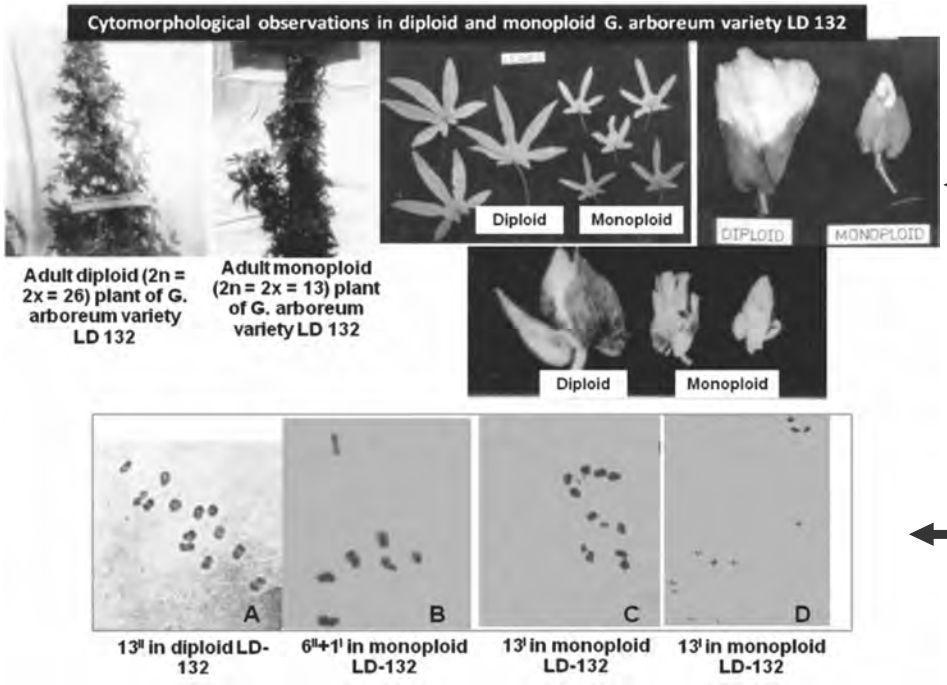
Sr.No.	Species	Variety	Aspect studied	Reference
A).	Spontaneous			
1	*G. arboreum*	LD 132	Report	Curr. Sci. 1977, 46, 349-350
2			Morphology	Natl. Acad. Sci. Letters, 1979, 2, 317-319
3			Cytology	Caryologia, 1980, 33, 393-400
4	Basic Chromosome Number			Curr. Sci., 1984, 53, 40-42

Utilization of haploids to obtain interspecific hybrids as a short cut : Haploids of tetraploid cottons are found crossable with *G. anomalum* (2n=2x=26, $B_1B_{1;}$ Kalyanaraman, *et. al.,*1954), *G. thurberi* (2n=2x=26 D_1D_1, Kalyanaraman, *et. al.,* 1955) and *G. arboreum* (2n=2x=26 A_2A_2; Kalyanaraman, *et. al.,* 1957). Since their hybrids are fairly fertility, they can be either useful in direct crossing with cultivable Asiatic diploid (2n=2x=26 AA) *G. herbaceum* (A_1A_1) or *G. arboreum* or crossing with tetraploid (2n=4x=52) *G hirsutum* ($A_hA_hD_hD_h$) and *G. barbadense* ($A_hA_hD_hD_h$) after doubling of their chromosomes.

CONCLUSION

Gossypium arboreum (2n=2x=26,A_1A_1) is a secondary polyploid evolved with the loss of one chromosome at diploid level 2n = 14-1=13 with subsequent duplication of the entire aneuploid chromosome giving rise to 2n = 26 (4b-2) chromosomes (Caryologia, 1980, 33 (3), 393-400 and Curr. Sci., 1984, 53(1): 40-42).

Cytolmorphological studied of the monoploid (2n=x=13, A_1) of *G. arboreum*, L (2n=2x=26,A_1A_1) Caryologia, 1980, 33 (3), 393-400)



(Nat. Acad. Sci. Letters, (September 1979); 2(9): 317-319;

Caryologia, 1980, 33 (3), 393-400

Earlier hypothesis

14

14 – 1 = 13

13 x 2 = 26

2b – 2

New hypothesis

7

7 x 2 = 14 – 1

13 x 2 = 26

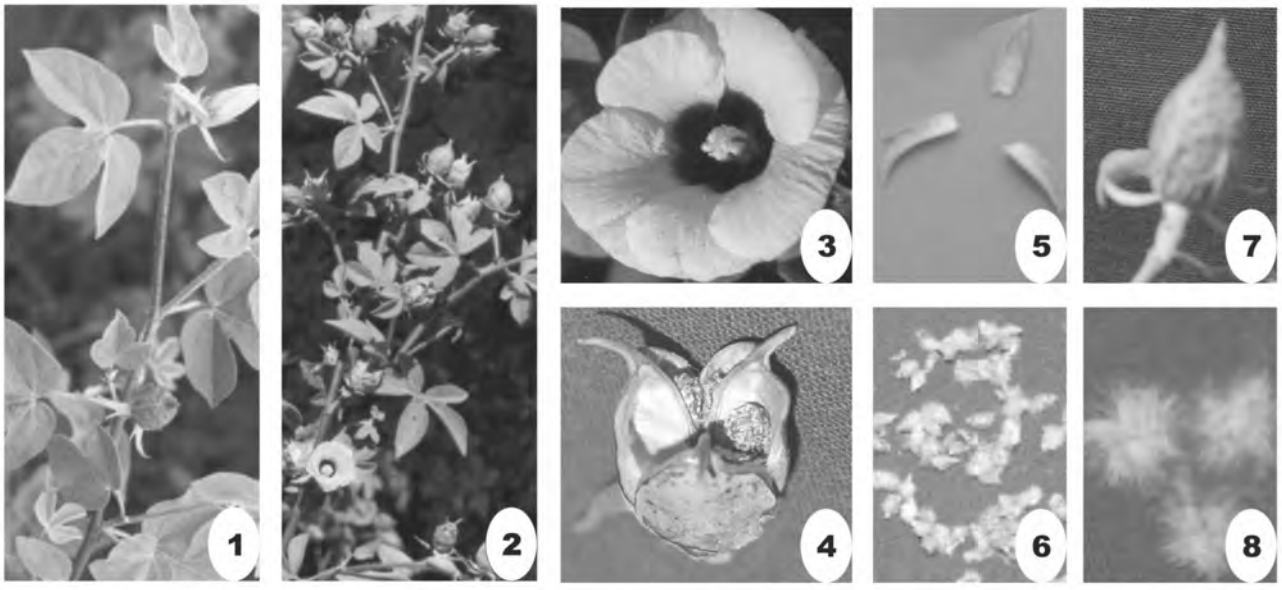
4b – 2

F_1 hybrids obtained utilizing haploids 2n = 2x = 26 (A_1D_h) and wild/ cultivated species of *Gossypium*.

Sr.No	Species with chromosome number and genome constitution	Reference
1	*G. thurberi* (2n = 2x = 26, D_1D_1)	*Cytologia,* 1981, 46 (1 &2): 291-299.*Indian J. Bot.,* 1982, 5(2), 120-122
2	*G. hirsutum* (2n = 4x = 52, $A_1A_1D_1D_1$)	J. *Maharashtra Agric. Univ.,* 1981, 6(3), 254-255
3	*G. anomalum* (2n = 2x = 26, $B_1^1B_1^1$)	*Indian J. Genet.*, 1982, 42, 144-149
4	*G. arboreum* (2n = 2x = 26, $A_1^1A_1^1$)	*Indian J. Genet.*, 2003, 63, 137-142
5	*G. barbadence* (2n=4x=52, $A_2^1A_2^1D_2D_2$)	*Indian J. Genet.*, 2003, 63, 175-177
6	Haploid x raimondii	*Indian J. Genet.*, 2003, 63, 319-324

Interspecific hybridization in *Gossypium* L. : Hybridization between species is resorted for securing genes or gene combination that is not normally available within the limits of species. Further improvement in certain characters through transgressive breeding is also possible. The interspecific hybridization was undertaken among cultigens and wild types of *Gossypium* is mostly on the hybridization between tetraploid (2n=4x=52) x diploid (2n=2x=26) species. Such transfer is possible with difficulties and breeding programme is comparatively lengthy because most F_1s being triploids are sterile, they can be backcrossed to cultivated tetraploids to obtain plants with 2n=4x=52 chromosomes having desired character combinations only after doubling its chromosome complimen.

Wild *Gossypium anomalum*: a unique source of fibre fineness and strength.



Morphological features of the African wild *Gossypium anomalum* Waw. & Peyr. *(Gossypium anomalum)* (2*n* = *2x* = *26; B1B1). 1, Branch at flowering; 2, branch at boll maturity; 3, pink flower with dark red petal spot; 4,*mature/burst boll; *5, narrow bracts; 6, seeds with brown fuzz; 7, matured boll with narrow bracts, and 8, brown,*scanty lint strongly adhered to seed coat. (*Curr. Sci.,* **99 (1) :** 158-71).

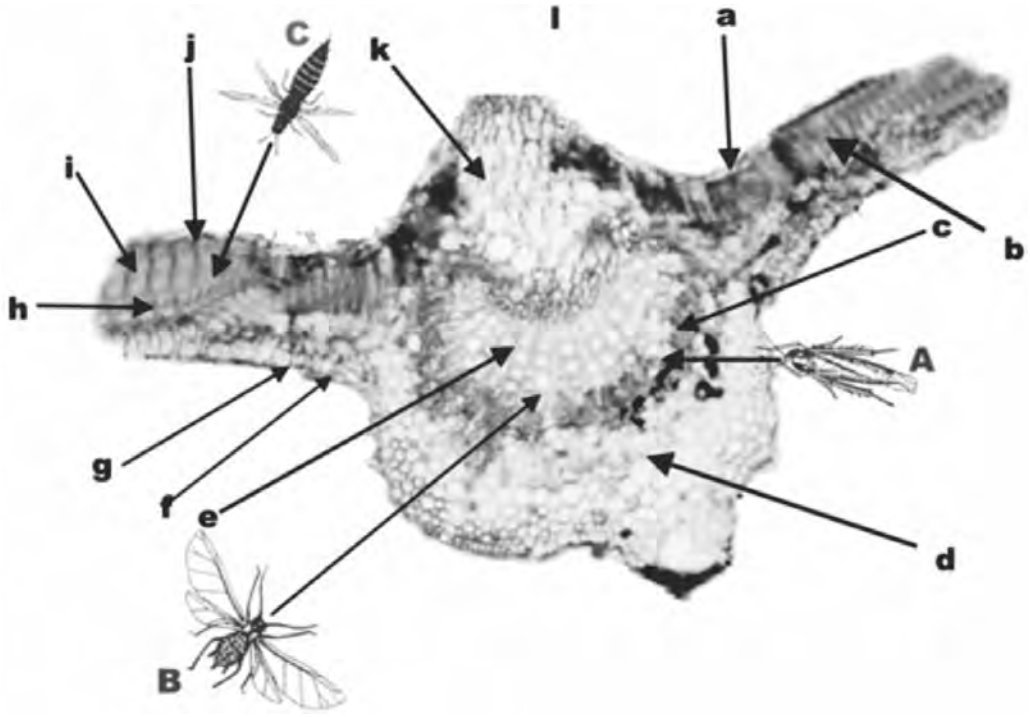
Introgression of high fibre strength and fineness from wild *G. anomalum* to cultivated cottons

Analysis of fibre properties in F_2 (amphidiploid G.arboreum x *G. anomalum) x G.hirsutum) x G.barbadnse.*

A) Variablity

S. No.	Characters	Mean	Minimum	Maximum	SD +/-	CV %
1	Fibre strength (g/t)	23.99	17.60	29.60	3.15	13.12
2	2.5 per cent Span length (mm)	31.06	25.50	35.80	2.52	8.10
3	Micronaire	2.97	2.40	5.10	0.48	16.20
4	Uniformity Ratio (%)	47.61	43.00	54.00	2.76	5.80

Introgression of Leaf anatomical characters confering resistance to multiple sucking pests from wild *Gossypium anomalum* to cultivated cottons.



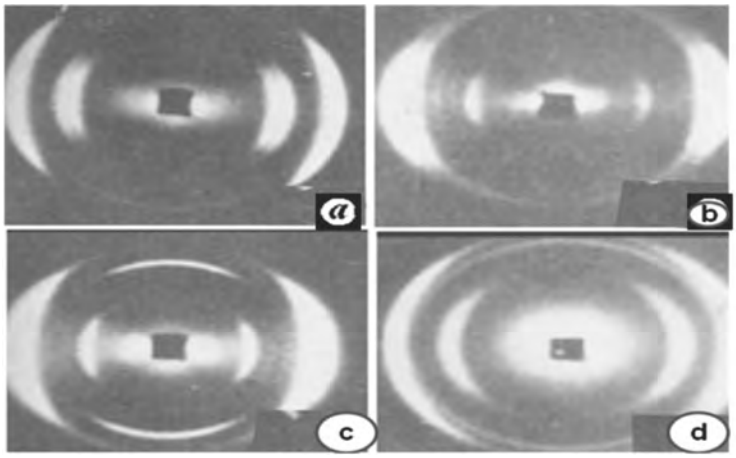
B) Fibre quality parameters of selected plants

Plant Number	2.5 per cent Span length (mm)	UR (%)	Micro-naire	Tenacity (g/t)
1	30.80	50.00	2.70	28.40
3	30.00	47.00	2.70	27.20
4	27.90	47.00	2.50	21.50
5	34.10	46.00	3.40	27.20
7	35.70	48.00	2.60	28.80
8	31.50	51.00	3.10	26.20
31	29.10	54.00	3.50	25.10
32	26.30	50.00	5.10	21.00
102	32.80	47.00	2.80	29.60
17	33.00	45.00	2.70	22.20
19	34.40	45.00	2.60	24.00
36	30.80	52.00	3.00	23.50
48	31.90	46.00	2.70	23.20
47	32.50	49.00	3.60	25.80
27	31.80	49.00	3.30	25.60
60	30.50	43.00	2.40	18.00
68	28.00	49.00	2.90	21.90
84	34.00	44.00	2.80	23.80
79	32.10	47.00	2.80	27.10
76	35.80	48.00	2.90	22.60
29	28.90	49.00	2.80	25.00
33	29.20	45.00	2.40	18.80
95	32.60	44.00	3.30	24.10
100	30.50	43.00	2.80	22.50
117	33.20	46.00	3.00	23.30
67	29.50	44.00	2.80	20.80
52	28.40	52.00	3.30	29.50
66	31.80	48.00	2.90	21.20
71	27.70	49.00	2.90	22.10
54	30.70	48.00	2.80	26.80
103	25.50	52.00	3.00	17.60
128	31.40	47.00	2.90	21.70

Transverse section showing anatomical structure of matured leaf of *Gossypium hirsutum L. cotton and feeding habit of major* sucking pests of cotton. a, Cuticle; b, Spongy tissues; c, Phloem; d, Parenchyma; e, Xylem; f, Lower epidermis; g, Stomata; h, Mesophyll; I,palisade layer; j, Upper epidermis; k, Collenchyma, and l, Sclerenchyma.A, Jassid; B, Aphid, and C, Thrip. CURRENT SCIENCE, VOL. 99, NO. 1, 10 JULY 2010, 58-71.

The nature of inheritance and gene action involved for the control of different morpho physiological traits, anatomical and biochemical parameters responsible for tolerance/resistance to sucking pest complex and bollworms were estimated using 'generation mean analysis'. Both additive and dominant gene actions were found significant in the inheritance of these traits. Duplicate epistasis for all morpho-physiological characters and complementary epistasis for chlorophyll and sugar content was predominant. Correlation studies indicated significant negative association of anatomical and biochemical traits with sucking pest complex except jassids. Four F3 progenies of the cross were promising for seed cotton yield, improved fibre properties and tolerance to sucking pest complex and bollworms. Cytological studies in the progenies indicated almost normal meiotic behaviour and pollen formation (Amolic,2005, Amolic, *et al.,* 2008).

Unique combinations of high fibre length, strength, and fineness in F_2 derivatives of Trispecies* cross *of cotton (Gossypium L. spp.) X-Ray Crystallographic study.,

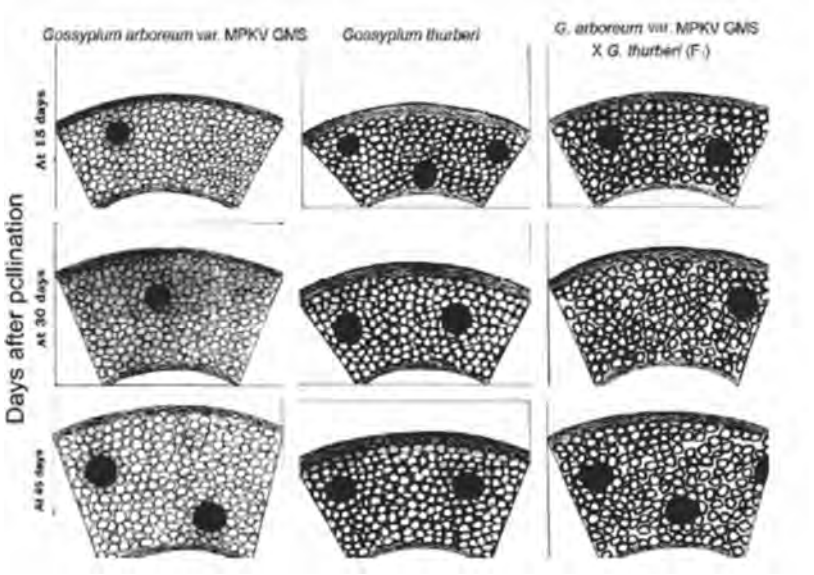


(Ph. D. Thesis- Jagtap, P.K., 2007. *Identification of molecular markers for fiber strength in segregating populations of interspecific crosses of Gossypium spp.;* **J. Indian Soc. Cotton Improv. (August, 2009), 34 (2): 96-106 and CURRENT SCIENCE, 10 JULY 2010,VOL. 99, NO. 1, 58-71**

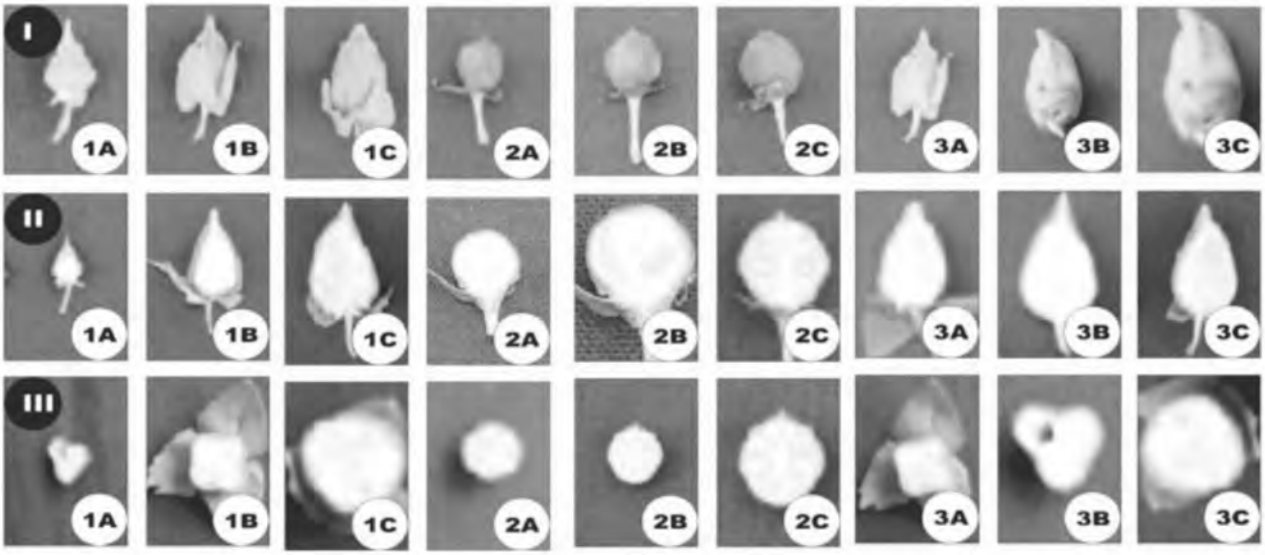
X-ray crystallography of parents, F1 interspecific hybrid and its selected F2 plants with X-ray diffraction radial scanning. Internal cotton fibre structure showing cellulose deposition and microfibrils. *a, Fibre sample of Gossypium anomalum b, Fibre sample of 2 (Gossypium arboreum × Gossypium anomalum). c, Fibre sample of Gossypium barbadense RHCb 001. d, Fibre sample of F1 interspecific* hybrid of tricross 2 (*Gossypium arboreum × Gossypium anomalum) ×Gossypium barbadense.*

Introgression of pink bollworm resistance from wild *Gossypium thurberi* Tod. to cultivated Gossypium *arboreum* L., cotton: pre-breeding efforts

Flower bud, flower and boll of *Gossypium thurberi* Tod (2n=2x=26; D_1D_1)



Drawing of cross sections sketches of boll rind *Gossypium arboreum* var. MPKV GMS, *Gossypium thurberi* and their hybrid at 15, 30 and 45-day-old bolls after flower opening



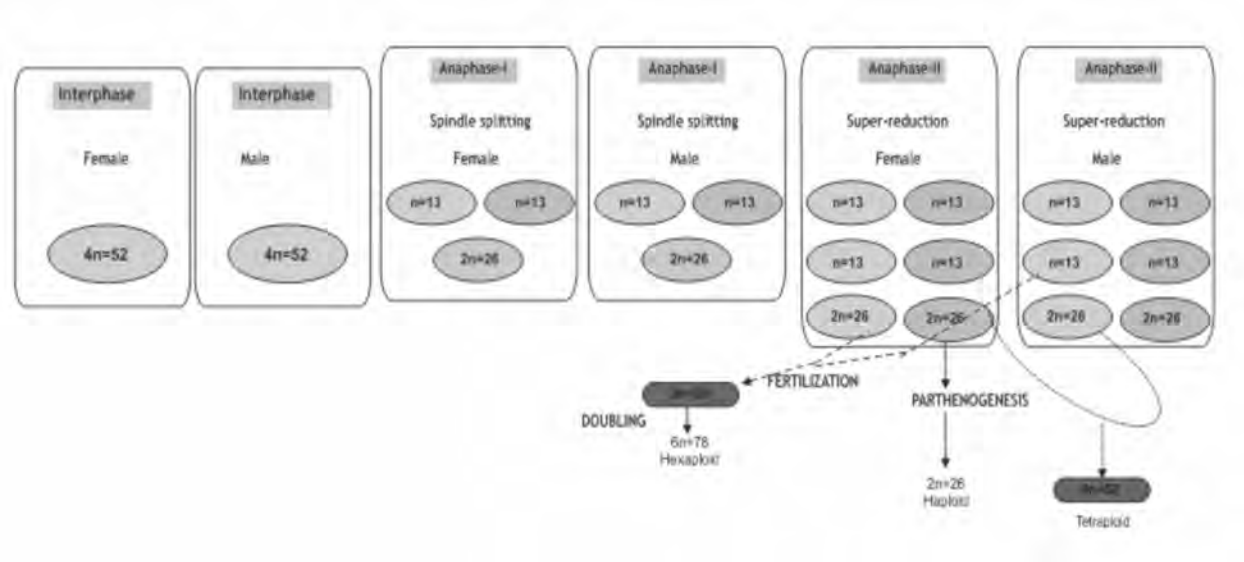
Observation recorded at A: 15, B: 30 and C: 45-day-old bolls after flower opening. 1-3, Bolls of 1: *Gossypium arboreum* var. x MPKV GMS (P_1); 2: *Gossypium thurberi* (P_2); 3: F_1 (*Gossypium arboreum x Gossypium thurberi*). I-III, I, Bolls of different ages; II, Longitudinal, and III, Cross section of the 15, 30 and 45-day-old bolls after flower opening.

(M.Sc. Thesis: *Patil J.M.* (2007) Inheritance studies in inter specific cross of cotton. & Curr. Sci., August 2009, 97(4), 558-564).

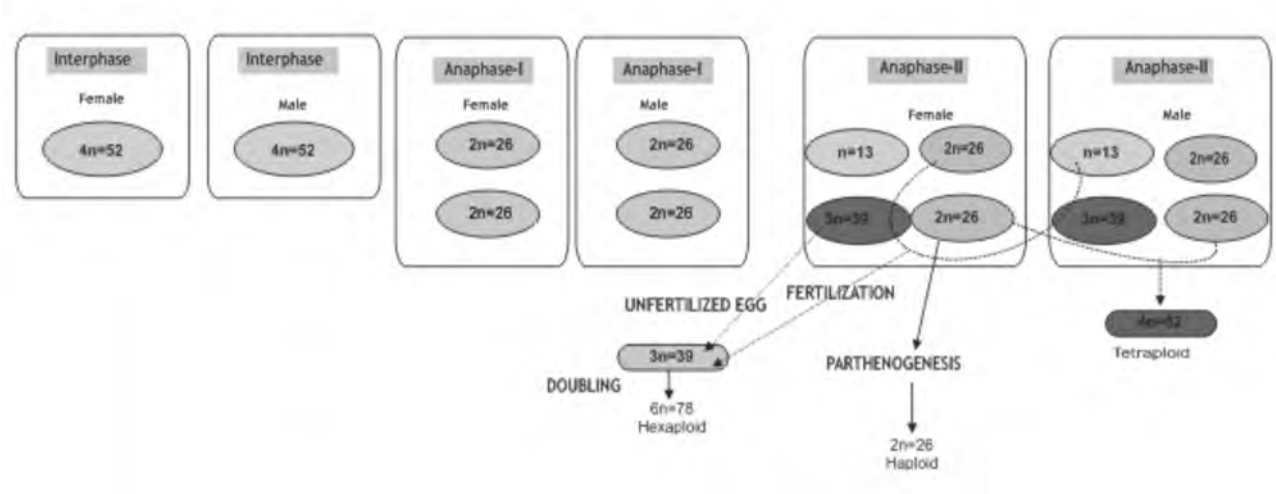
Occurrence of triploid (2n=3x=39) plants in cultivated tetraploid (2n=4x=52) cotton

SN	Species	Variety	Aspect	Reference
1	*G. hirsutum*	Laxmi	Report	Sci. & Cult., (July 1980); 46(7): 259.
2	*G. hirsutumG. barbadense*	Laxmi MCU-5	Cyto-morphology	Cytologia, 1982, 47,555-563.

Characterization of Progenies with Different Ploidy Multi-Genomic Background (Nevaskar et.al.,2013, Plant Breeding, 132, 211–216.)



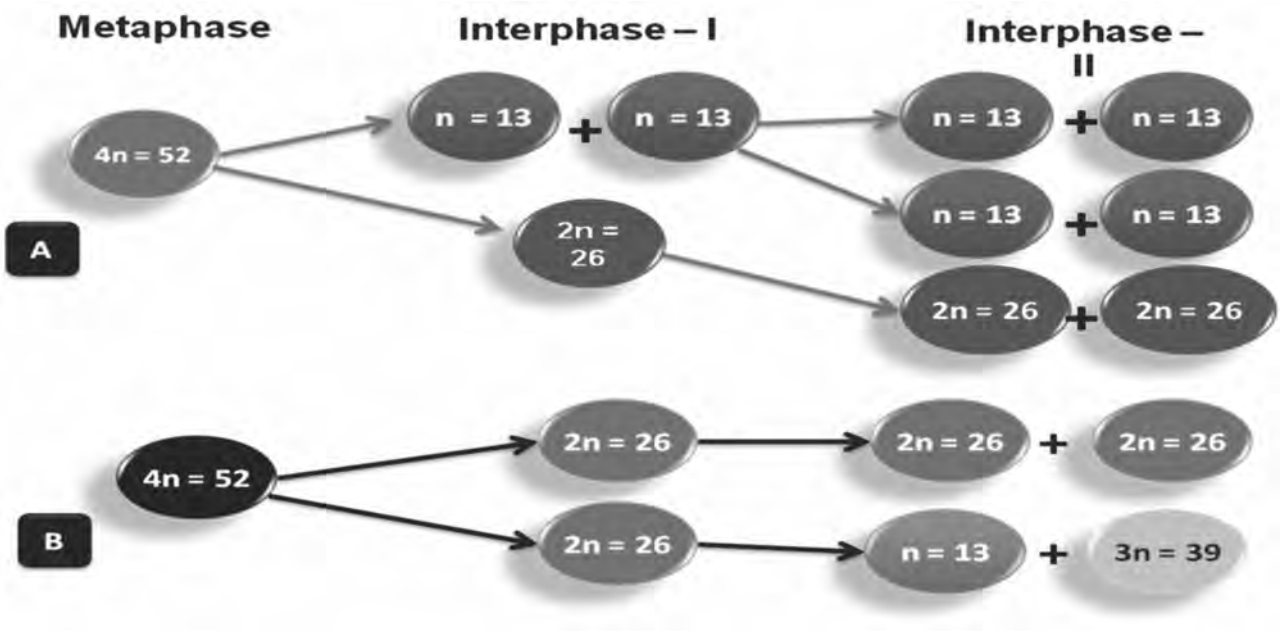
Abnormal first meiotic division in Anaphase-I due to tripolar spindle formation results in three nuclei with n+n+2n chromosome numbers. Further subdivision at Anaphase II leads to four haploids and two diploid microspores



First meiotic division is normal; misdistribution of n+3n in one of two nuclei in Anaphase-II results in 1 haploid (n=13), one triploid (3n=39) and two diploid (n=26) microspores

Possible origin of hexaploid and haploid plant in normal tetraploid due to abnormal chromosomal separation at anaphase I & II.

Origin of triploid by spindle splitting phenomenon

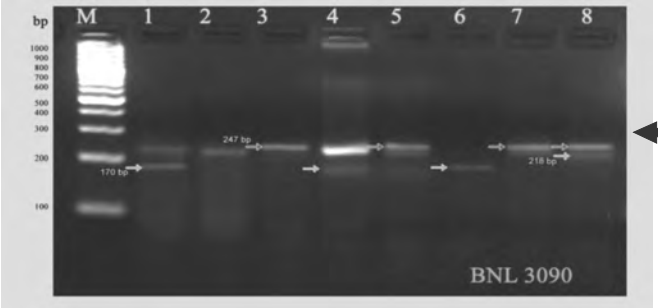


Molecular confirmation of interspecific hybrids

The list of hybrids confirmed by molecularanalysis

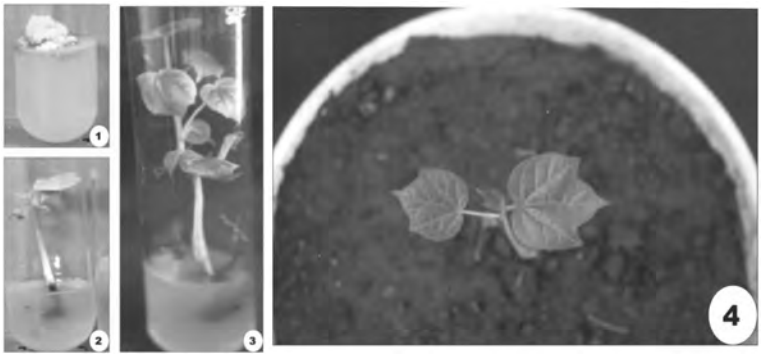
Sr.No	Hybrid	Reference
1	*G. arboreum* x *G. stockii*	*Caryologia*, 2004, 57, 171-175
2	Amphidiploid A_1 of *G. arboreum* x *G. capitis-viridis* – F_1 F_2 generations	*Cytologia*, 2004, 69, 367-369
3	*Wild species and primitive races*	*Int. J., Trop. Agri., 2006, 24, 177-186*
4	*G. arboreum x G. thurberi*	*Caryologia, 2007, 60, 379-388*
5	*G. arboreum x G. anomalum*	*Cytologia, 2008, 73, 213-219*
6	*G. arboreum x G. anomalum*	*J. Indian Soc. Cotton Improv., 2009, 34, 155-163*

Gel profile on BNL3090 microsatellite marker analysis



M- Marker; Lane 1: *Gossypium arboreum* var. MPKV GMS; Lane 2: Amphidiploid / Doubled F1 (*G. arboreum* x *G. anom*alum); Lane 3: Haploid; Lane 4: F_1 (*G. arboreum* x *G. anom*alum); Lane 5: Hexaploid; Lane 6: *G. anomalum*; Lane 7: Number One tetraploid; Lane 8: *G. hirsutum* var. JLH168

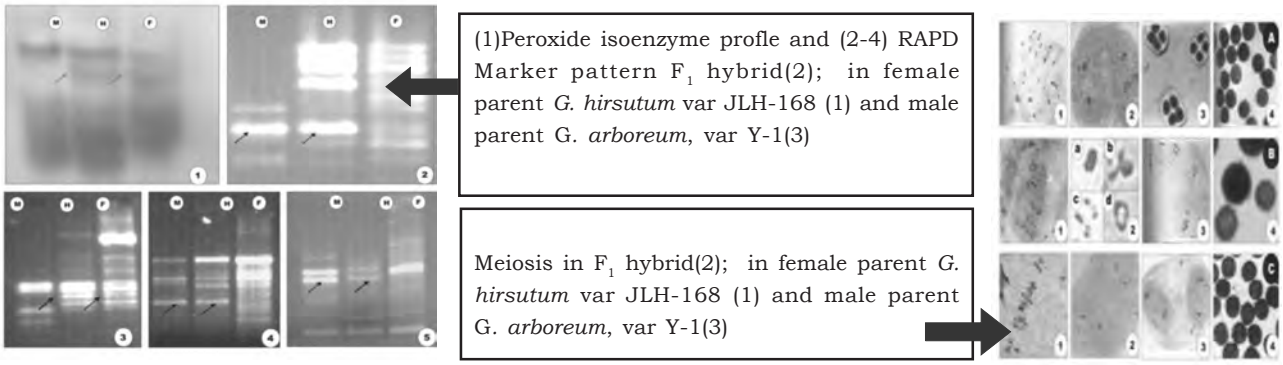
In ovulo embryo cultured hybrid between *Gossypium hirsutum* and *Gossypium arboreum*: hybridity confirmation.



1-3.In-vitro regeneration from ovules

1. Growth of calli 35 DAP fro ovules cultured I M-4 medium
2. Germination of ovule after 75 days
3. 90 days old seedling
4. 110 days old seedling after hardening

[Expression of morphological Characters in the F_1 hybrid(2); in comparison with female parent *G. hirsutum* var JLH-168 (1) and male parent G. *arboreum*, var Y-1(3). A-Leaf size ,shape and petiole length(a) ventral side and(b) dorsal side; B=Flower bud before two days of opening showing size, shape and serration of bracteoles; C= Androecioum showing filament, anther colour, and shape and dehiscence (1&3). And Non dehiscence (2).D= Flower colour, and shape showing+ and – of petal spot and its intensity; E= petal spot and its intensity= size, shape and serration of bracteoles; G= Fully developed boll size, shape and coverage of bracteoles,H= Opened boll containing seed cotton (1) and (3) and without seed cotton(2).]

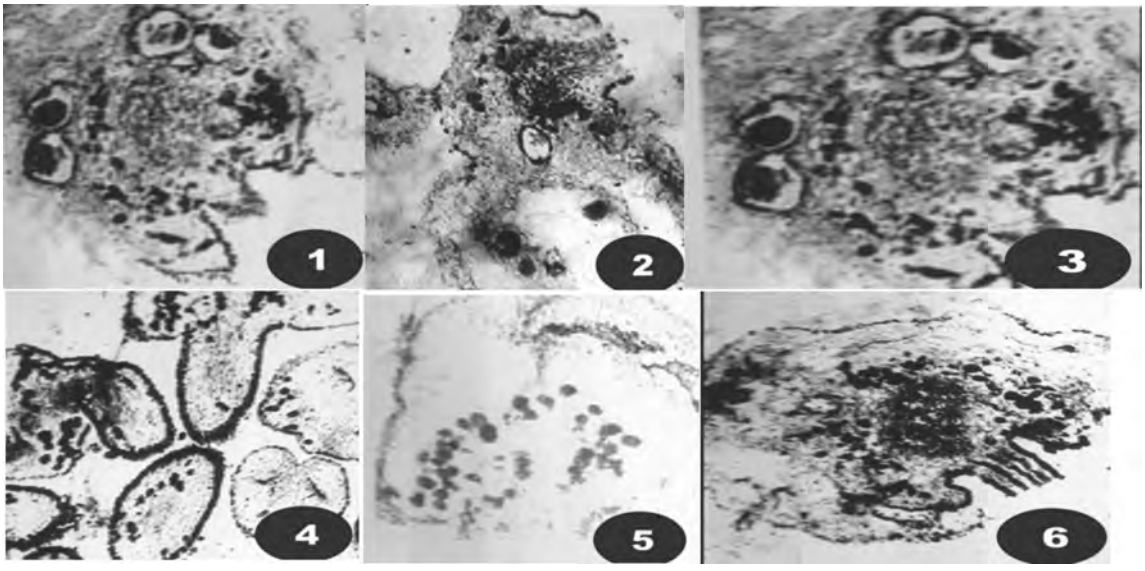


(1)Peroxide isoenzyme profle and (2-4) RAPD Marker pattern F_1 hybrid(2); in female parent *G. hirsutum* var JLH-168 (1) and male parent G. *arboreum*, var Y-1(3)

Meiosis in F_1 hybrid(2); in female parent *G. hirsutum* var JLH-168 (1) and male parent G. *arboreum*, var Y-1(3)

Ph. D Thesis, Pardeshi, S.V.(2004). Interspecifichybridisation between Gossypium hirsutum x G. arboreum, G. sturtianum, G. australe through ovulo-embryo culture. **&** J. Cotton Res. Dev., (July, 2007); 21(2): 131-139 **Anther and pollen development in cotton male sterile stocks, haploids and their parents.**

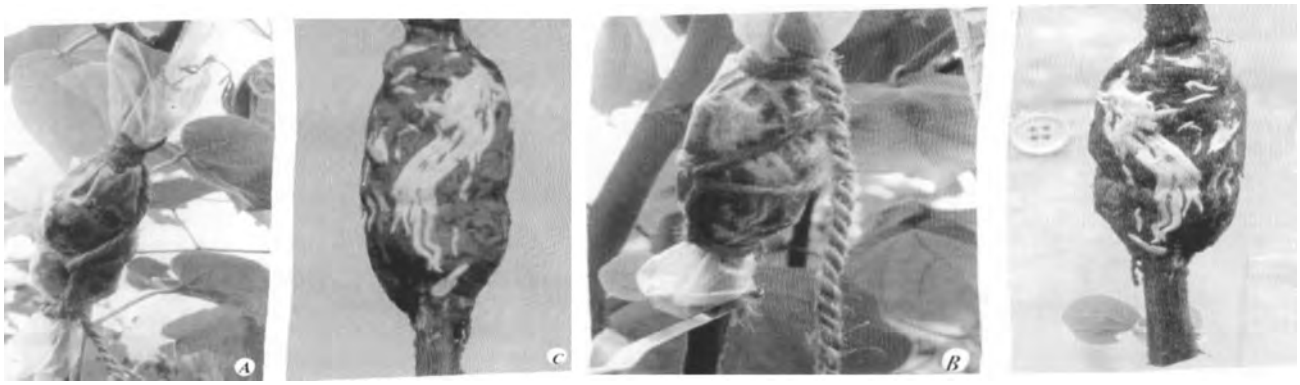
SN	Material	Reference
1	Male sterile stocks of Gossypium hirsutum, L. cotton	J. Maharashtra agric. Univ., (May 1981) : 6 (2) : 159-161
2	Haploids and their parents of G. hirsutum cotton	Proc. Indian Acad. Sci. (Plant Sci.). (October, 1982);91(5) 409-416.
3	Triploid and tetraploid plants in G. hirsutum	Indian J. Hered. (January-December, 1984). 16 (1-4): 23-29.



LS of anthers (1). G. hirsutum Genetic male Sterile "Gregg" (2) G. hirsutum Normal male Fertile "JLH-168" (3) G. hirsutum Triploid "Laxmi" 4-6 G. hirsutum Cytoplsmic Male Sterile lines

***Gossypium raimondii*: a new source of fertility restorer for cytoplsmic male sterility of Gossypium hirsutum, L**

Vegetative propagation of haploids in cotton by air layerage. Air layering in cotton – A tool to propagate wild species of Gossypium and their interspecific hybrids.



J. Maharashtra agric. Univ., (May 1977); 2(2): 163-164. *J. Plant. Biology*. December 2002, 29 (3): 331-334

During cource of investigations different new techniques used/reported are as under

Sr.No.	Technique	Reference
1	Vegetative propagation of haploids in cotton by air layerage.	*J. Maharashtra agric. Univ.,* **2(2):** 163-64.
2	Detection of haploids- stomata- chloroplasts	*Sci. & Cult.,* 1982, **48,** 75-77.
3	Differential stain	*J. Maharashtra Agric. Univ.,* 1980, **5(2),** 166-67
4	Chromosome doubling in a haploid	*Sci. Cult.,* 2000, **66,** 207-08
5	One line theory for fixation of heterosis	Sci. Cult., 2000, 67, 383-85
6	Overcoming cross incompatibility in Gossypium species	*J. Pl. Biol.,* 2002, *29,* 33-38; *J. Cot. Res. Dev.,* 2002, **16,** 111-24; *Indian J. Biotech,* 2004, **3,** 29-36
7	Vegetative propagation air layring	*J. Pl. Biol.,* 2002, **29,** 331-34
8	Overcoming interspecific incompatibility	*Curr. Sci.,* 2003, **84,** 1510-12
9	Rapid multiplication of GMS G. arboreum plants	*Sci. Cult.,* 2003, **69,** 299-301
10	Conservation of wild Gossypium species	*Indian J. Genet.,* 2003, **63**, 353-54

Introgression of characters from wild to cultivated cotton

SN	Trait /Character	Source	Reference
1	Pink bollworm resistance	G. thurberi	Cur. Sci., 2009, **97,** 558-64
2	High fibre length, strength, fineness	G. anomalum	*J. Indian Soc. Cotton Improv.,* 2009, **34**, 96-106
3	Unique fibre fineness	G. anomalum	*Cur. Sci.,* 2010, **99,** 58-71

Use of wild species for hybridization with cultivated *Gossypium* species

Species with chromosome number and genome constitution	Reference
With *G. arboreum* (2n=2x=26; A_1A_1)	
G. *stocksii* (2n=2x=26; E_1E_1)	*Indian J. Pl. Genet. Reso.* 2002, 15, 213-218
G. thurberi(2n=2x=26; D_1D_1)	*SABRAO J. of Breed. Genet.,* 2003, 35, 71-79

Species with chromosome number and genome constitution	Reference
Hybrids between wilds x wilds species of *Gossypium*	
G. davidsonii (2n=2x=26; $D_{3\text{-}d}D_{3\text{-}d}$) *x* *G. anomalum*(2n=2x=26; B_1B_1)	*SABRAO J. Genet.,* 2003, 35, 43-56
With *G. arboreum* (2n=2x=26; A_1A_1)	
G. anomalum (2n=2x=26; B_1B_1)	*Nucleus,* 2003, 46, 138-146
With *G.hirsutum* (2n = 4x= 52, $A_1D_1A_1D_1$)	
G. *arboreum* (2n=2x=26; A_1A_1)	*J. Cotton Res. Dev.,* 2007, 21, 131-139

Intergeneric hybrid obtained

Species with chromosome number and genome constitution	Reference
G. hirsutum (2n = 4x = 52, A_h A_h D_h D_h) x *Hibiscus panduraeformis* (2n=24)	Euphytica, 1980, 29,323-330.

Different Phenomenon reported

Sr. No.	Phenomenon	Reference
1	Parthenogenesis	J. Maharashtra Agric. Univ., 1982, 7(2), 187
2	Triploidy	Cytologia, 1982, 47(3/4), 555-563
3	Basic chromosome number	Curr. Sci., 1984, 53(1), 40-42
4	Petaloidy	Sci. & Cult., 2003, 69, 75-76

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Resistance management in *Bt* cotton

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Abstract: Transgenic *Bt* cotton crop that express cry proteins for the control of bollworms have been commercialized across the cotton growing countries of the world. Growers have enthusiastically embraced crops genetically improved to express *Bt* proteins for insect control because they provide excellent protection from key damaging insect pests around the world. *Bt* crops also offer superior environmental and health benefits while increasing grower income. However, Insect Resistance Management (IRM) strategies have strengthened pest management systems by identifying appropriate insecticides so as to delay resistance, ensure effective control of target pests, and conserve naturally occurring biological fauna for enhanced sustainability of ecosystems. Insect resistance management for *Bt* cotton is of great importance, because the development of insect resistance to Cry toxins threatens the longevity of the effectiveness *Bt* technology. IRM is an important part of stewarding this valuable technology. IRM needs for *Bt* cotton vary among countries because of differences in pest status, pest biology, farming practices, economic status of the farmers, level of literacy among cotton growers and experiences. In countries like India, dominated by smallholding farmers, the scale and diversity of the agricultural systems poses significant challenges for IRM but also presents opportunities. Appropriate IRM strategies should consider alternative crop and non-crop hosts as sources of unstructured refuge, particularly for highly polyphagous pests such as the cotton bollworm, *H. armigera.* Whereas, for monophagous and oligophagous pests like PBW and SBW structured refuge with non *Bt* cotton crop should be considered coupled with strategies like gene pyramiding and high dose with a eagles eye on the changes in levels of susceptibility through base line resistance monitoring.

Insect control traits introduced into plants using modern biotechnology methods have shown high economic value across the globe. An assortment of crops expressing different *Bacillus thuringiensis* (*Bt*) proteins has been commercialized for insect control and additional products are under development. Growers have embraced crops (*i.e.* maize, cotton, potato and rice), genetically improved to express different *Bt* proteins (*i.e.* Cry1Ab, Cry1Ac, Cry1F, Cry1A.105, Cry2Ab, mCry3Aa, Cry3Bb, Cry3Aa, Cry34/35, Vip3A), as they provide excellent protection from key damaging insect pests in global regions (ISAAA, 2009). There is a history of safe use of these proteins, both for the environment and human health (Betz *et al.,* 2010). Recent evaluations have shown that these traits provide economic value to adopting countries through increased grower income (Brookes and Barfoot, 2009). The continuing value of this technology can be enhanced through appropriate stewardship, such as insect resistance management (IRM) plans, that can prolong trait efficacy against the target insect pests. Since the commercial release of transgenic *Bt* cotton, incorporating a gene for a highly specific insecticidal protein from *Bacillus thuringiensis* in 1996 in US, many countries including India have adopted the technology. *Bt* cotton has found favour with farmers in many parts of the world and the area under *Bt* cotton has been increasing year after year, currently at 0.5 m.ha. As a result there has been tremendous reduction in overall use of

insecticides (Patil *et al.,* 2008) and better environment in cotton growing regions. *Bt* cotton appears to yield 20 to 40% more than non *Bt* cotton in field trials. The yield will depend upon the insect infestation levels and several other factors. Cost benefit ratio will be the main criterion for the farmers to accept *Bt* cotton (Patil *et al.,* 2008).

It is expected that any competitive biological system would respond to high level of selection pressure by mechanisms that would either avoid or mitigate it. Random genetic changes that keep happening in a population of insects might include resistance alleles at very low frequency, which can rapidly increase when challenged. *H. armigera* has already developed resistance to many potent insecticides, especially to pyrethroids (McCaffery *et al.*, 1989; Armes *et al.*, 1996; Kranthi *et al.*, 2001, Fakrudin *et al.,* 2004). There is also an indication that mechanisms of detoxification for different insecticides do overlap (Vijaykumar and Patil, 2005). In this context, wide spread use of *Bt* cotton and other *Bt* crops has to be considered. Like with chemical insecticides, *H.armigera* has a potential to develop resistance to Cry toxins under field conditions due to continued selection pressure, throughout the crop growth period, if proper resistance management tactics are not implemented. So far there is no field resistance observed for *Bt* cotton. However, wide geographic variation in susceptibility of *H. armigera* to Cry1Ac toxin has already been reported in India (Gujar *et al.,* 2000; Kranthi *et al.*, 2001; Fakrudin *et al.,* 2003; Jalali *et al.*, 2004), China (Wu *et al.*, 1999) and in Australia (Liao *et al.,* 2002). The ability of lepidopterans to develop resistance to Cry toxin under laboratory conditions was demonstrated well before the commercial release of *Bt* transgenics (Tabashnik *et al.*, 1994; Gould *et al.*, 1995; Kranthi *et al.,* 2000) and subsequent studies in laboratory and on field collected larvae do point to this fact (Vijaykumar and Patil, 2005).

Development of resistance to Cry protein is a concern, which is being addressed by evolving different management strategies. Besides refuge crop, there are other ways of resistance management *viz.*, gene pyramiding, application of insecticides and biorationals at a critical stage of the crop and other IPM strategies for delaying resistance build up in the insect population or to keep it at strategically low levels. Predictions based on a stochastic model with input parameters for Indian conditions, Kranthi and Kranthi (2004) have estimated that it required *H.armigera* 11 years to reach resistance allele frequency of 0.5. Semi-dominance for resistance to the toxin, 40 per cent cotton area under *Bt* cultivars, very low initial frequency of resistance allele were some of the assumptions and refuge crop at 20 per cent would delay resistance development by two more years. In fact, 11-13 years is a good period for resistance to hold under modern agriculture. However, resistance development in insects is real and it has to be 1) managed with a sound IRM strategy, 2) *Bt*technology used as a component of IPM, 3) limited use of insecticide molecules in case of partial or complete failure of Cry toxin and 4) gene pyramiding whenever necessary. As of now, ETL based application of chemical pesticides in *Bt*cotton is recommended once after 90 DAS (Kranthi *et al.*, 2004) or 1-2 times (Rajanikantha and Patil., 2004) based on ETL.

Cross resistance : Accurate prediction and management of resistance requires information on cross-resistance characteristics of the insecticide employed in *Bt* crops. Reports on the cross resistance between various categories of Cry toxins are available (Akhurst

and liao, 1996; Zhao *et al.*, 2000; Liao *et al.*, 2002) and our studies indicate negative cross resistance between cry toxins and chemical pesticides employed in cotton (Vijaykumar and Patil, 2005).

IRM challenges :

- Pest/crop considerations are country specific
- Adoption/distribution of a *Bt* crop will affect selection intensity on insect pests
- Practicality and feasibility of the IRM strategy will affect implementation of the plan
- Cost of plan implementation to growers, industry, and the government/regulatory agencies
- Accountability for adherence to IRM plan
- Geography of region where IRM plan is implemented
- Crop production practices in region where IRM plan is implemented

The nature of insect resistance: Genetics studies indicated that Cry1Ac resistance allele in *H. armigera* was incompletely dominant. Cry1Ac resistance allele in *H. armigera* in India was incompletely dominant (Kranthi, 2003). Bioassays of *P. gossypiella* (Liu *et al.*, 1999) and *P. xylostella* (Tang *et al.*, 2001) on Cry1Ac transgenic plants showed that the effective dominance (D_{ML}) was completely recessive. However, results of Kranthi (2003) indicated that the effective dominance (D_{ML}) in *H. armigera* was semi-dominant. Resistance to Cry1Ac was reported to be autosomal and inherited as incompletely recessive trait in *H. armigera* (Akhurst *et al.*, 2003), *Plutella xylostella* (Sayyed *et al.*, 2000; Tabashnik *et al.*, 1997), *Heliothis virescens* (Gould *et al.*, 1992) and *Pectinophora gossypiella* (Liu *et al.*, 1999). In general, is inherited as an incompletely recessive trait. Insect populations often have natural genetic variation affecting response to a toxin, with some alleles conferring susceptibility and others conferring resistance. Alleles conferring resistance are typically rare in insect populations before the populations are exposed to a *Bt* toxin, with empirical estimates often close to one in a thousand (Downes *et al.,* 2009, Huang *et al.,* 2009).

Factors affecting the development of resistance : There are several factors that increase the rate at which insecticide resistance is generally developed. Some factors are related to the insect population itself: species with higher reproductive rates, shorter generation times, greater numbers of progeny, and larger, more genetically varied local populations develop a large resistance population more quickly. Whether the genetic basis of insect resistance is dominant or recessive is also of importance (Wearing and Hokkanen, 1995). Other factors are dependent upon the insecticide. Resistance develops more rapidly to more persistent insecticides; their staying power in the environment increases the chance that susceptible individuals are exposed to the toxin and die, thus not passing on their insecticide-susceptible traits to the next generation. This selects more strongly on resistant insects because only the resistant insects thrive. By similar logic, frequent application of non-persistent insecticides has the same effect. Insect populations with little immigration into the gene pool of new, non exposed susceptible individuals also develop resistance more readily. Populations that have in the past been exposed to an insecticide with a mode of action similar to that of a new insecticide are quick to develop resistance to the new toxin. This phenomenon is known as cross resistance (Pimentel and Burgess, 1985).

Factors to be considered to formulate IRM strategies: IRM strategy devised for *Bt* cotton that includes: suitable spatial and temporal expression of the *Bt* protein; some form of refuge for susceptible pest insects; use of alternative control measures (placement of *Bt* crops into an integrated pest management context); monitoring and remedial action plans; and the development of subsequent products with different insecticidal mechanisms. For any particular *Bt* crop in any particular country, however, it is necessary to understand the resistance risk and what sort of IRM options may be feasible by collecting information on the agricultural system, the biology of the target pests, and the behavior of growers, and generating local information on product performance. This helps to define the precise IRM tactics that can and should be used.

Accurate resistance monitoring requires evaluation of insect field populations on *Bt* crops as well as from other sources including non *Bt* host plants. Sampling and testing of target pest insects surviving on or near *Bt* crops is essential for early detection of field -evolved resistance (Tabashnik *et al.,* 2008). Failure to sample such insects favors underestimation of the frequency of resistance, which can postpone detection of resistance and is contrary to the primary purpose of resistance monitoring. Although most research on *Bt* toxins focuses on physiologically based resistance, behavioral changes can also cause resistance by reducing exposure to a toxin (Onstad 2008).

The following factors should be considered :

- The cropping system
- Biology of target insects
- Grower behavior and attitudes and
- Product performance

Insect resistance management programs for *Bt* cotton have been developed, implemented, and refined to delay insect resistance development. Experts from industry, academia and regulatory agencies have worked together to design and implement effective resistance management plans that are crop and region-specific. Such plans, based on the best available science, typically include:

- Baseline measurements of target insect susceptibility to the *Bt* protein prior to commercialization,
- On-going monitoring of target pests for changes in susceptibility after commercialization,
- An adequate supply of susceptible insects to mate with any resistant insects, achieved through appropriate practical programs such as: Grower-planted refuges/Natural or cultivated alternative hosts,
- Grower education programs to enhance awareness of general IRM concepts and specific grower responsibilities,
- The newest generation products express additional *Bt* genes or other insecticidal gene combinations with multiple modes of action that increase the durability of these technologies.

Universal strategies for resistance management :

Pest monitoring and surveillance: The data is collected by various agencies. However, there is no standard guide line for collection and use of such data. Monitoring of resistance should also be done for pink, spotted bollworm and tobacco caterpillars. The information should be shared with crop entomologist and technology generators.

Resistance Monitoring: The objectives of resistance monitoring are

- To document resistance if developed in any region
- To measure and identify resistant genotypes
- To provide an early warning of an impending resistance problem
- To determine changes in the distribution or severity of resistance
- To make the need based insecticide recommendations
- To test the effectiveness of IRM strategies.

Monitoring and product stewardship: Determinations of baseline susceptibility to the *Bt* protein expressed in the *Bt* crop and subsequent post commercial monitoring for insect resistance are an integral part of all IRM plans. For every commercialized biotech *Bt* crop, the baseline susceptibility of each of the target pest species to the relevant *Bt* protein is to be assessed as part of the overall IRM strategy.

Second generation *Bt* cotton (Bollgard II):Recently , Monsanto, USA released Bollgard II for commercial cultivation in the USA, Australia, India and other countries. Bollgard I was based on event 531 of the Cry 1 Ac, whereas, Bollgard II contains Cry 2Ab as event 15985, in addition to Cry 1Ac. The stacked genes are expected to increase the spectrum of toxicity to a wide range of lepidopteran insects to include *Spodoptera litura*, which is known to survive Cry 1Ac. The strategy is good for resistance management, but would have been still better if it would have in a simultaneous release, rather, than one toxin first, followed by, the mixture containing the toxin. Similar deployment of conventional insecticides has led to multiple resistance.

Seed rate: The SAU is recommended higher seed rate and is between 650-750g/acre. The same should be adopted for better plant stand. The need is to educate farmers not to use non *Bt* seed for gap filling.

Optimum hybrids: Large number of hybrids can create many problems in sustaining productivity. Many hybrids released are not having good yield potential which results in lower productivity. There must be minimum standard of productivity and fiber quality for release of any hybrid. The data of insect and disease should be generated under high pest population and hybrids with high degree of tolerance to sucking pests should only be stand in future.

Refugia: 'Refuge' is one of the most favored resistance management options that have been preferred all over the world. The refuge strategy is designed to ensure that *Bt*susceptible insects will be available to mate with *Bt*resistant insects, should they arise. The offspring of these mating will be *Bt*susceptible, thus mitigating the spread of resistance in the population. The strategy relies on several conditions. Mainly the alleles should be recessive or the expression levels of the toxin in transgenic plants should be adequate to kill heterozygous larvae. Other conditions include that mating is random, frequency of resistance alleles is rare and that there is no fitness cost associated with the resistance allele. However, for the Indian situation it may be required to redefine the parameters to strengthen the concept of refuge.

Current refuge requirements for *Bt* cotton

- A 95:5 external refuge, where 5 percent of the acres of non *Bt* cotton must be

planted for every 95 per cent of the acres of *Bt* cotton. In this option, the non *Bt* cannot be treated for target pests. The refuge must be within one-half mile of the biotech field.

- An 80:20 external refuge, with 20 per cent of non *Bt* acres for every 80 per cent of *Bt* acres that can be treated with conventional sprays for target pests (excluding *Bt*k sprays). The refuge must be within one mile of the biotech field.
- An embedded 95:5 refuge, where the non *Bt* crops are situated in blocks within the *Bt* field. The crops may be treated with conventional sprays (aside from *Bt* sprays) that kill target pests but only when the *Bt* field is also being treated.

Refuge in bag (RIB) : To ensure refuge compliance as a proactive measure towards insect resistance management (IRM), a new approach has been developed wherein *Bt* and non *Bt* seeds are pre-mixed in a recommended proportion and made available to farmers in the same packet or bag. This technique is referred to as 'refuge in a bag' (RIB) in USA, where it is deployed for the *Bt* corn products and in India as 'refuge in mixed bag'. For the sake of uniformity and better clarity, the name 'refuge in seed-mix' (RISM) appears to be more appropriate and is used in this note (Manjunath, 2007).

The objective of RISM in *Bt*cotton is to ensure that non *Bt* plants are randomly distributed among *Bt*cotton plants in a field in a pre-decided proportion. The presence of such randomly distributed non *Bt* plants may raise certain concerns such as:

- potential movement of the later instar larvae of *Helicoverpa armigera* from non *Bt* to *Bt* plants, thus causing crop damage and
- exposure of such migrant larvae to sub lethal dose of *Bt* protein, thereby increasing the chances of resistance development.

These concerns appear valid, but their actual impact needs to be examined in the Indian context from a practical perspective, especially in view of certain data available elsewhere on the adverse fate of the *Bt* fed later instar larvae and also that RISM is considered as a practical strategy towards IRM in pursuit of preserving a remarkable technology like *Bt*cotton (Manjunath, 2007).

The role of alternative hosts in IRM, particularly in developing countries : Of particular importance in defining refuge strategies for developing countries is the role of alternative hosts of the target pests. If the target pests are utilizing a wide variety of alternative host crops, and they are not being controlled using *Bt* on these other hosts, then structured refuges for *Bt* crops may not be necessary under these conditions; the alternative host plant species will act as an adequate source of refuge for the *Bt* crop (Siegfried *et al.,* 2006; Sivasupramaniam *et al.,* 2007). In these cases, both cropping practices and the degree of polyphagy of the target insect species will be important. For IRM approaches based on alternative host plant species to be effective, several conditions should be examined :

- the target pest species must utilize multiple host plant species that overlap in both space and time;
- the performance on the different host plant species must be comparable to allow the different alternative hosts to produce sufficient susceptible insects at the right time to interbreed with any resistant insects emerging from the *Bt* crop;

- the distribution of these different host plant species must overlap at a sufficiently fine scale and consistently enough to act as a functional refuge in all relevant areas; and
- the pest insects must move freely between the different host plant species.

Low expression : The option of low expression has been ruled out as a IRM strategy globally as this is likely to result in inadequate and unacceptable levels of pest control. However, it is expected to reduce selection pressure that allows the survival of susceptible alleles to thrive in the form of heterozygous insects.

High expression : High toxin expression is a favored option from the stand point of efficient pest control. It favours reduction of heterozygous individuals but banks on susceptible strains from refuges to maintain an overall susceptibility. The success of the high dose strategy depends on rare and recessive or partially recessive resistance alleles (Huang *et. al.,* 1999). But, temporal changes resulting in a reduction in toxin expression can help heterozygous individuals to overcome the toxin and thus spread resistance alleles. Additionally, heterozygotes must have no advantage over insects homozygous for the susceptibility allele, or the development of resistance can be hastened (Curtis, 1981). At the given state of art of cotton-transformation technology in India, it may take a few years to achieve a high expression in straight varieties so that the technology can look really robust.

Regulating expression: Bollworms rarely feed on leaves. They prefer fruiting parts. Theoretically it sounds attractive to consider the option that tissue specific promoters are used to ensure only fruiting parts produce the toxin so that they are protected while the insect pest is encouraged to feed on leaves, which may at the most result in insignificant yield losses. But, conversely it is possible that young larvae survive on leaves, grow into older instars capable of surviving the toxin levels in fruiting parts, and thus cause yield losses. The toxin regulating choice however, has never reached any practical stage in any of the transgenic development programmes till date. But, because, toxin expression in almost all commercial *Bt*cotton crops is known to decline after a certain stage, it may be useful to consider the use of promoters (example Late Embryogenesis Abundant (LEA) promoters) to ensure that the toxin is produced even at late stages in the crop life. Most resistant management approaches aim at reducing selection pressure in order to delay resistance development. Reduction in selection pressure can be brought about by either a temporal regulation of toxin expression as an insect inducible response or a tissue specific expression.

Gene pyramiding : Gene stacks offer an attractive option of delaying resistance especially if two or more genes are available with high toxicity levels having independent modes of action and are do not have cross resistance. The strategy also helps in increasing the efficacy of pest control and in turn reducing refuge area with enhanced toxicity spectrum. Scientists are currently working on plants with "pyramided" or "stacked" proteins — plants that express more than one protein. One example is BOLLGARD II, a Monsanto product that expresses two *Bt* genes, that is approved for commercial use. Using such a method, insects would have to possess resistance alleles for each individual protein in order to survive – a very unlikely event. Diverse

"*Bt*" proteins have been identified, permitting a number of possible gene combinations.

Spatial and temporal transgene deployment : Two different transgenic varieties each incorporating the individual toxin gene, to be grown either as mosaics or rotated one after another every year. Though mosaics have never been shown to be practically useful in resistance management programmes, rotations have certainly been used throughout for insecticide resistance management. Thus rotation of two or three different transgenic crops alternatively one after the other each year would cause a reduction in selection pressure due to each of the single toxins, and thus delay resistance development.

Restricted planting times: This avoids late maturing and therefore minimizes the crop's exposure to high densities of *H. armigera*.

Hectare restrictions: Growers are restricted to a maximum of 400 ha or 30 per cent of the total cotton grown per farm unit and in situations where the *Bt* area is less than 400 ha but exceeds 30 per cent of the total cotton grown there are additional refuge requirements.

Spray limitations: To ensure that adults emerging from refuge crops have not already been selected for resistance to *Bt* protein, no *Bt* sprays are allowed on any refuge crops for the entire season. Any management action that negatively affects the population of *Helicoverpa* spp. is regarded as an 'insecticidal' action. Inter-row cultivation destroys pupae and is not allowed in refuge fields unless the same action is carried out in the associated *Bt* cotton fields. Food sprays are not permitted in refuge crops. Food sprays will increase the levels of beneficial insects which will decrease the *Helicoverpa* spp population, acting as a form of 'biological' insecticide.

Trap crops: In Central Queensland, *Helicoverpa* spp pupae produced late in the season do not remain in the soil but emerge within 15 days of pupating. Trap crops can be employed to attract any adult *Helicoverpa* emerging after the cotton has been cut out. After the cotton is harvested the trap crops should be destroyed, removing the food for the larvae which will then die.

Control of volunteers: The presence of *Bt* cotton volunteers within a conventional cotton crop imposes further selection pressure for *Bt* resistance. Conventional cotton volunteers within an *Bt* cotton crop are also of concern. Growers are required to remove volunteers as soon as possible from all fields planted with *Bt* cotton following conventional cotton and from all fallowed and conventional fields following *Bt* cotton.

Combining insect control methods: The next general approach to resistance management combines elements of both old and new ideas. It is assumed that resistance is less likely to evolve to two control methods simultaneously than to only one method. Thus, with two methods, resistance to the combination will be delayed more than using each individually in a temporal or spatial arrangement. Using two or more control methods at a time is like having insurance in case one of them begins to lose efficacy. Similar to this is a method of heightening effectiveness of a toxin by using a high dosage like having one dose as the first control method and a second dose as the other, though the underlying theory is

different.

Compliance: Grower compliance with refuge and IRM requirements is a critical element for resistance management and significant non compliance may increase the risk of resistance. However, it is not known what level of non compliance would compromise the risk protection of current refuge requirements. To minimize the effects of non compliance, the EPA considers that it may be necessary to develop a broad compliance strategy as part of the IRM strategy.

Use of economic thresholds and integrated pest management (IPM): Integrated pest management (IPM) that uses *Bt* cotton as one of the component is the ultimate option of IRM including

- cultivation of sucking pest tolerant *Bt* genotypes genotypes,
- seed treatment (Imidacloprid 70 WS or Thiomethoxam 70WS @ 5-7g/kg),
- inter cropping with cowpea, soybean and blackgram,
- stem application of acetamiprid or thiomethoxam or imidacloprid at 40 DAS,
- avoidance of broad spectrum organophosphates as early season sprays,
- identify and use attractive synchronous alternate host crops for *H. armigera,* which could be used as intercrop or trap crop refuges,
- use alternate genes that do not share common resistance mechanisms as that of Cry1Ac, in transgenic plants either in rotation or alternation or mixtures.

New insights into resistance management strategy for bollworms in Indian conditions: In north India the key pests species are pink bollworm (a monophagous pest), spotted (Oiligophagous) and American bollworm (polyphagous). For pink and spotted bollworm the carry overpopulation is from cotton to cotton only as cotton is grown as a monocrop in north India due to which any crop other than cotton cannot be grown as refugia for all the three bollworms. Hence, the structure refugee of cotton hybrid with same growth pattern as sprayed or unsprayed is the only viable option. Whereas, for south and central India chickpea is sown along with cotton that can act as refugia. But for management of spotted bollworm okra is suggested. However, pink bollworm is coming up as a serious pest and refugia need for the same can only be from non *Bt* cotton. The possibility of using okra as refugia crop is still not very sound approach. Option can be given for use of cotton/okra/pigeon pea as refugia. Insecticide spray for management of resistance population: This is a good strategy and needs validation by all institutes.

CONCLUSION

Biotechnology can be used for or against our advantage in resistance management. Further research can tell us which way to go. *Bt* based transgenic crops represent the state of art in pest management. So for the profitability of *Bt* technology monitoring resistance of bollworms to Cry1Ac in *Bt* cotton, essential management strategies to slow the resistance, significance of alternative host plants in resistance management and development of suitable newer genotypes by gene pyramiding which express higher levels of Cry protein in terms of their resistance to target pests and proper IRM strategies and spraying of suitable insecticides at later stages of crop growth to kill resistance

population would help to overcome the resistance problems in *Bt* cotton. In the meanwhile, *Bt* producers and growers should be conservative and careful in creating, enforcing, complying with, and monitoring current resistance strategies. *Bt* is nearly an ideal pesticide and the loss of its use would be an extremely unfortunate occurrence. It is therefore necessary to enrich and preserve the life of such an excellent eco-friendly pest management strategy. Development and execution of appropriate resistance management strategies are imperative to ensure sustainability of the technology. Each of these elements must be assessed with a specific focus on the local conditions and agricultural practices that should be considered in the development of an IRM plan, because no matter how detailed the scientific information, these local conditions are critical for successful IRM deployment.

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Genetic transformation in cotton (*Gossypium hirsutum*)

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Cotton (*Gossypium hirsutum*) is one of the most important sources of fiber in the world. More than 80 countries are involved in its cultivation and a large number of industries spread in 150 countries are producing products based on cotton fiber. Cotton cultivation area in India is the largest in the world andis second in production next toChina.Cotton is a major cash crop in India whose production and market controls finances and lives of millions of small farmers and workers linked to cotton industry. The fiber is used in textiles, medical and other industries where short fuzz on the seed, which is called linters, provides cellulose for making plastics, explosives and other products. Cottonseed oil is third after soya and canola oils produced in the world and isused in cooking and products like soap, margarine, emulsifiers, cosmetics andpharmaceuticals, rubber etc. As fiber consists of more than 90 per cent cellulose, a long carbohydrate polymer, waste cotton can be enzymatically degraded into glucose and can be fermentedto produce ethanol in future.

Cotton is one of the most vulnerable crops for several insect pests and abiotic stresses. More than 60 per cent of the pesticides in India were used for cotton previously. Further our productivity s one of the lowest in the world around 500 kg/ha. One of the ways to improve the cotton is to increase the productivity atpar with other countries so that land for cotton cultivation can be utilizedto grow other important crops to produce other vegetable oils and pulses that India is at present dependent on imports. It is well known that the most important contributor for the Indian success of production from less than 100 kg to more than 500 kg/ha represently is release of *Bt* cotton in the country, which occupies presently more than 90 per cent of cultivated area. However, despite being the largest producer in the world with 25 per cent of land cultivated, our productivity to area is the least in the world and contributes only 18 per cent, while China produces 25 per cent with lesser acreageto total world production(TCCI report).This is one of the major challenges India still is facing. Although BT cotton is a great success for bollworm control, cotton is still susceptible to several other smaller pests, which are now emerging as major pests and destroying cotton. One of the recent examples is the emergence of a previouslysmaller pest- whitefly, destroying BT cotton causing 60 per cent loss in Punjab this season. The gene pool to develop whitefly resistant cotton seems very limited or not good enough to use conventional approach of plant breeding methods. Combining with lack of germplasm for most of the biotic and abiotic stresses, there is a constant and pressing need for incorporating genes from related/un related plant species and other organisms in to cotton through genetic engineering to overcome these stresses.

One of the ways to incorporate genes is genetic transformation of cotton plant. Cotton is one of the most difficult crops amenable for transformation methods; often the methods are very laborious and time consuming. So far

several methods have been developed and tried for generatingtransgenics in several labs all over the world. In most of the labs Coker 310 or 312 were used, as they are more amenable to tissue culture and regeneration methods. Either particle bombardment for direct gene incorporation in to the plant parts or *Agrobacterium*-mediated transfer of genes has been used for cotton transformation with different antibiotics such as Kanamycin as selectable agents.

Different methods of cotton transformation

Ex plant transformation : The foremost cotton transformation procedure was based on using ex-plants from cotyledonary leaf pieces and hypocotyl sections from germinated seedlingsto co-cultivate with *Agrobacterium*. The selection medium generally contains antibiotics like kanamycin thatgive rise to transformed callus;which is then converted to somatic embryos and finally into plants. It is a multi-step process involving labour-intensive work over a 10–12 month period starting from co-cultivation of *Agrobacterium* with cotyledonary or hypocotyl ex-plants, followed by production and maintenance of several hundreds of calli derived from independent transformation events on a selection medium, induction of embryogenesis in each callus line and the development and germination of somatic embryos into normal plantlets. In this procedure, the transformation efficiencies are generally low due to the low frequency of embryogenesis and the difficulty in germination of transformed embryos (Firoozabady *et al.,* 1987; Umbeck *et al.,* 1987, 1989). Also, lack of proper root development among the germinated embryos lowers the transformation frequency still further (Chaudhary *et al.,* 2003). Due to long culture periods the chances of somaclonal variation, infertility is high(Trolinderand Goodin, 1987).However, the first transgenic cotton plants created using the particle gun method was reported by Finer and McMullen (1990) where embryogenic suspension cultures of *G. hirsutum* L. cv. Coker 310 were transformed using particle bombardment.

Embryonic suspension cultures of cotton were used for particle bombardment to transform where it is easy to produce large amount usable cells in a short time and the regeneration rate is high when combined with multiple bombardments, the transformation rate is high Rajasekaran *et al.*,, (1996, 2000). However, suspension cultures are genotype-dependent, only a few varieties can be regenerated into plants. Though they used cryo-preserved suspension cells to avoid abnormalities, it was not a practical method for laboratories not equipped with cryopreservation facilities.One of the best methods is using embryogenic calli for transformation with *Agrobacterium* (Leelavathi *et al.,* 2004) which reduced time several months taken for the normal transformed calli to turn into embryogenic calli which in turn regenerated in to normal plants through metabolic stress easily (Kumria *et al.,*2003). Use of embryogenic calli as explants other than leaf or hypocotyl for transformationsaved time to convert the callus to embryogenic calli and it was made possible to obtain transgenic plants within 6 months. Later several improved methods increasing the transformation efficiency of the technique further were reported in other varieties of cotton other than Coker 310 (Jin *et al.*,, 2005, Khan *et al.*,,2010, Wu *et al.*,, 2011). However, unless the regeneration through embryogenic calli is well established, this technique is not feasible in all the varieties cotton.

Table 1. Important genes transformed in cotton.

Gene	Function	Refrence/Year
Cry1Ac (Bollgard)	Resistance against lepitoderan pests	Monsanto 1996
Cry1Ac+Cru2Ab (Bollgard II)	Resistance against lepitoderan and *Spodoptera spp.* pests	Monsanto 2002
Cry1Ac +Cry1F (Widestrike)	Resistance against *Spodoptera spp.* pests	DowAgrosciences 2004
Vip3A+Cry1Ab (VipCot)	Resistance against *Spodoptera* spp. pests	Syngenta
Cat and nptII	neomycinPhospho transferaseII marker	Umbeck *et al.*,1987
nptII and OCS	neomycinPhospho transferaseII marker	Firozabady 1987
hpt	Hygromycin resistance	Finer and McMullen 1990
Cry1Ac,Cry1Aband nptII	Resistance against lepitoderan pests	Perlak *et al.,* 1990
nptII and tfda	2-4 D resistance	Bayley *et al.,* 1992
Fiber specific FbL2A promoter driving phaB,phaC and phaC	Fiber thermal properties were altered	Rienhart *et al.,* 1996
Fiber specific E 6or FbL2A promoter driving phaB,phaC	Fiber thermal properties were altered	John and Keller 1996; Chowdhury and John 1998
AHAS gene	Resistance to herbicides imidazolinone and sulfony urea	Rajasekaran *et al.,* 1996
CP4-EPSPS	Resistance to herbicides Glyphosate	Nida *et al.,* 1996; Chen *et al.,* 2006
Mn-SOD, APX,GR	Chilling induced inhibition of Photosystem II reduced	Kornyeyev *et al.,* 2001,2003a,b
SAD-1 and FAD 2-1	Higher Stearic acid and oleic acid level	Liu *et al.,* 2002
Endochitianse	Protection against Aleternaria and Rhizoctonia	Emani *et al.,* 2003
CryV	Resistance against lepitoderan pests	Leelavathi *et al.,*. 2005
FAD-2	Seed oil with higher oleic acid	Sunilkumar 2005
D4E1	Resistance against several fungal pathogens	Rajasekaran *et al.,* 2005
NHX1	More fibre and biomass under salt stress	He etal 2005
ACA	Resistance against aphids	Wu *et al.,* 2006
Delta-Cadiene synthase	Reduction in gossypol level	Sunilkumar *et al.*,2006
SPS	Improved fibre quality	Haigler *et al.,* 2007

Transformation of shoot apex : Since embryonic axes can regenerate into plants without a callus intermediate, using shoot apexs for gene modificationoffers an attractive alternative simpler genotype-independent and faster transformation method. This method is used to generate transgenics either through particle bombardment or *Agrobacterium* mediated transformation. Shoot apex, or shoot meristem areas from water imbibed seeds or very young seedlings are used as explants. Later, McCabe and Martinell (1993) reported a successful transformation of cotton by using excised embryo axes as explants through bombardment method.Chlan *et al.,* (1995), Keller *et al.,* (1997) also reported the successful transfer of a foreign gene into cotton by bombardment method.The advantage of using the embryo meristem as an explant is that it allows genotype independent transformation and the relatively rapid recovery of transgenic progeny (Christou, 1996; John 1997). The disadvantage of using embryonic meristems is that the preparation of large number of shoot tip meristems is an extremely tedious, labor intensive task, which involves the surgical removal of leaf primordia to expose the meristem, followed by thecareful excision of meristem explants from imbibed seeds. Also, the

stable transformation rate is very low (0.001 to 0.01 per cent) and mostly true transgenics are identified in screening of second generation.

Transgenic cotton plants have been produced by biolistic bombardment of organized shoot-tip meristems (McCabe and Martinell 1993; Chlan *et al.,* 1995; Keller *et al.,* 1997). Satyavathi *et al.,* (2002)transformed three Indian cultivars where they could recover transformed shoots from kanamycin selection medium. Nevertheless, it is a laborious method involving huge number of shoot tips.

***In planta* transformation :** Transformation techniques that evade tissue culture (Graves and Goldman, 1986) become important in recalcitrant crops such as cotton. Such *in planta* transformation techniques have also been standardized in other crops like, buckwheat (Kojima *et al.,*, 2000), mulberry (Ping *et al.,*, 2003), kenaf (Kojima *et al.,* 2004), soybean (Chee *et al.,*, 1989) and rice (Supartana *et al.,*, 2005) etc. In this method, the strategy essentially involves in planta inoculation of embryo axes of germinating seeds and allowing them to grow into seedlings *ex vitro*. These *in planta* transformation protocols are advantageous over other methods because they do not involve time consuming *in vitro*regeneration procedures and therefore the tissue culture-induced somaclonal

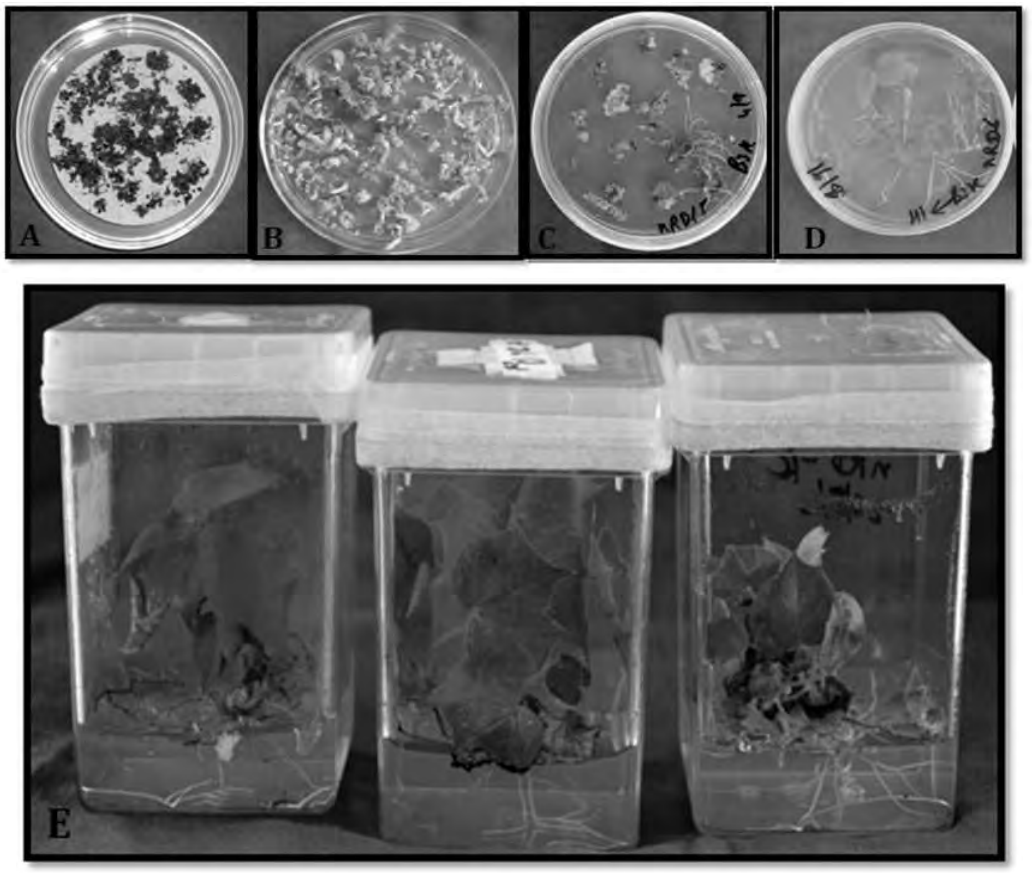

Fig. 1. Genetic transformation and plant regeneration. (A) Selection of transformed callus (B) Embryo induction in transformed callus (C) Embryos germination. (D & E) Plants regeneration, transfer of plantlet with good roots.

variations are avoided. Keshamma *et al.,* (2008) andAlkuddsi *et al.,* (2014) used meristems of seedlings to transform cotton. Alkuddsi *et al.,* (2014) followed similar method to transform Cry1Ac-Cry1Ec genes but without any confirmed results in T1 generation. While the method is independent of genotype, only in the T1 generation one can screen for transformants where huge number of seedlings have to be collected and it is a very tedious process for screening. For labs with limited greenhouse facilities and space this method can be very difficult to follow.

One of the more efficient germline transformation methods were reported by Chen *et al.,* (2014) where large numbers of mechanically isolated meristem explants of cotton were transformed using *Agrobacterium* containing GUS with spectinomycin as a selection marker. It was reported to be efficient and would be a valuable method if it can be reproduced in other laboratories and other recalcitrant species like legumes,woody species etc.

Pollen tube transformation : The pollen-tube pathway is distinct from the previous methods because it does not require *in vitro* based cell or tissue cultures. Flowering cotton plants are allowed to self pollinate. Pollen forms a tube when it is on stigma of a flower and reaches the ovule to fertilize the eggin ovule. Once this has occurred, the ovary, which contains the ovules, is exposed by removing the petals and a solution containing DNA carrying gene of interestis injected into the ovary. The DNA travels down the pollen tube to the ovule and is integrated into the genome of the developing cotton embryo (Zhou *et al.,*1983, Wang *et al.,*, 2004) through a mechanisms that is still not understood clearly. When the ovules mature into seeds, the seeds are planted and selected for successful transgene integration. In contrast, any cotton cultivar can be transformed using the pollen-tube pathway because regeneration is not required. In addition, the pollen-tube pathway does not require a marker gene for the selection process because other methods such as PCR, Southern blot analysis that can efficiently determine whether a plant is transgenic. More recently, TienZi *et al.,*(2010) reported better transformation efficiency of 0.46 to 0.93 per cent when they cut off stigma before pistil drip with *Agrobacterium* on the same day of flowering.

Chloroplast transformation : This method overcomes concerns of gene containment, low levels of transgene expression, gene silencing, positional and pleiotropic effects or presence of vector sequences in transformed genomes. Several therapeutic proteins and agronomic traits have been highly expressed via the tobacco chloroplast genome but extending this concept to important crops has been a major challenge; lack of 100 per cent homologous species-specific chloroplast transformation vectors containing suitable selectable markers, ability to regulate transgene expression in developing plastids and inadequate tissue culture systems via somatic embryogenesis are major challenges.

There is only one report from Kumar *et al.,* (2004) so far and no other laboratories reported chloroplast transformation in cotton.

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Prospectus of *Bt* cotton in Haryana

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Cotton is the most important commercial crop of India It provides employment to millions of people in various activities such as cultivation, seed production, marketing and industrial utilization. Cotton plays an important role in Indian economy in spite of severe competition from synthetics fibre in recent year. *Gossypium* includes 50 species, four of which are cultivated, 44 are wild diploids and two are wild tetraploid (Percival and Kohel, 1990). Out of the four cultivated species, *Gossypium hirsutum* L. and *Gossypium barbadense* L. commonly called as new world cottons are tetraploid (2n = 4x = 52), whereas, *Gossypium herbaceum* L. and *Gossypium arboreum* L. are diploids (2n = 2x = 26) and are commonly called as old world cottons.

Major cotton growing are China, India, United States, Pakistan, Brazil, Turkey, Australia etc. More than 90 per area of cotton is under *G. hirsutum*. Cotton crop suffers heavy losses (about 58%) in seed cotton yield mainly due to insects in North India. As this crop is attacked by more than 230 species of insects all over the world, however 10 –15 insects are considered important and six insects cause major yield losses (Ridgeway, 1984). The damage due to sucking pests is about 18.5 per cent and bollworm contribution is 30.3 per cent.

Cotton insects are divided into following two groups.

a. **Sucking pests**: - jassids, aphids, thrips, whitefly and leaf roller.

b. **Tissue feeders**: - American bollworm, pink bollworm, spotted bollworm, tobacco cutworm, stem weevil, ash weevil and red cotton bug.

Extent and nature of damage of bollworms

Bollworms: Spotted bollworms, pink bollworm and American bollworm damage the fruiting bodies and are known as the important bollworm pests of cotton. These bollworms cause reduction in yield and quality of seed cotton.

a. **Spotted bollworms (*Earias* spp):** *Earias insulana* and *E. vittella* are the two important species of spotted bollworms which damage cotton during vegetative as well as reproductive phases of the crop. Adults of *E. insulana* dominate over *E. vittella* in the drier parts of cotton growing areas. The bud infestation results in the flaring and shedding of buds. This pest adversely affects both yield and quality of cotton.

b. **Pink bollworm (*Pectinophora gossypiella*) :** The larva is creamy in first instars, white in the second, watery in the third and pink with dark brown head in the last instars. It feeds on buds, flowers and bolls. Flower parts and the developing seeds are the main food of the larvae. This pest causes quantitative as well as qualitative loss of yield, seed and lint.

c. **American bollworm (*Helicoverpa armigera*):** It is the most important pest of cotton. Under favorable weather conditions during 2001-2002 crop season the insect caused almost failure of the crop. However, under normal infestation conditions it can cause 20-40 per cent reduction in yield, in spite of usual pesticide use.

The boll worm complex *i.e.* pink boll worms, spotted boll worms and American boll worms resulted in heavy losses to Cotton crop and the crop could not be protected against these pests inspite of 8-10 sprays. In India *Bt* cotton was introduced to manage the bollworms particularly *American* boll worm as this pest developed resistance to insecticides and threatened the cultivation of cotton. In India *Bt* Cotton cultivation started in the year 2003 with the introduction of Bollgard I (Mon 531 cry IAC), after that Bollgard II (Mon 15985-Cry 1 Ac and Cry 2 A6) was released for cultivation in 2006, JK Agri genetics developed "Event I " and Nath Seeds GFM event (fusion gene-cry 1 Ac/cry 1 Ab). Since introduction more than 1000 *Bt* Cotton hybrids have been approved for cultivation based on these four events. During almost a decade after their release *Bt* gene provide effective control against boll worms complex during the complete crop duration and no report of emergence of bollworms as serious pests was observed.

Effects of *Bt* cotton

- A sudden rise in the hybrid cotton acreage.
- A phenomenal increase in the production and productivity of the country.
- Change in the farmers preferences for agronomic traits after introduction of *Bt*
- Significant reduction in pesticide usage for bollworms after introduction of the technology.
- A phenomenal shift in cultivation of *hirsutum* in the country after introduction of *Bt* technology and *arboreum* are almost on the verge of extinction.
- A significant shift in fibre class in the country. Shortage of short and medium staple cotton.
- More preference to big boll hybrids in the country after introduction of *Bt* technology.

Problems: Some problems have became more prevalent since the cultivation of *Bt* Cotton hybrids namely

- Sudden wilting of plants
- Increased damage of sucking pests particularly whitefly
- Threat of minor pests of cotton like mealy bug, myrid bug, aphids and thrips to emerge into major pests
- Resistance to *Bt* gene
- Low yield potential and narrow adaptability
- Improvement of fibre quality to meet the industrial needs

Biotechnological applications

- First generation products:
  (Towards reducing input cost)
  Bt cotton herbicide tolerance
- Second generation products
  (Improvement of output traits like fibre quality and abiotic stress)
  - Leaf curl virus disease resistance
  - Gene pyramiding
  - Transferring fibre quality genes

However, in spite of more than 90 per cent area under *Bt* Cotton hybrids and effective control of boll worms, the yields are declining for

that attention is required.

1. Ban on use of illegal *Bt* Cotton hybrids.
2. Screening of *Bt* Cotton hybrids suitable for different agro climatic conditions.
3. Sowing of refugia to harness the advantages of *Bt* Cotton technology for longer period.

Solution : Only genotypes with proven yield advantage, adaptability and proven fibre quality should be encouraged to make Indian cotton cost competitive, quality worthy and comparable with the international cotton.

Upcoming challenges

1. Changing weather pattern
2. Appearance of new pests and resurgence of existing diseases on the crop
3. Resistance management
4. To produce more from less for more
5. Hybrid seed production
6. Meeting industry needs

The cultivation of cotton has become costly affairs due to pest menace. Efforts on breeding for resistance or tolerance to insects in cotton were made during mid 1980's and early 1990's, but all such conventional efforts never resulted into a perfect cultivar which exhibited resistance to bollworm. Mean while the introduction of new engineering of biological molecules by manipulating the genetic makeup opened the new era for development of genetically modified plants. Transgenic cotton with resistance to lepidopteran insects has been released for cultivation in India in 2002. It has been observed that in a short span of period, there has been a remarkable adoption of this technology in India and it covered about 95 per cent of the total area under cotton. During this period, the yields and overall production has increased and India continued to maintain the largest area under cotton cultivation and second largest producer of cotton next to China with 34 per cent of world area and 21 per cent of world production, as a results became a net exporter of cotton from a being a net importer.

More than 300 *Bt* cotton hybrids has been recommended for North India of different seed compaies and each seed company make efforts to capture maximum share in seed market and farmers became in confusing state regarding the choice of proper hybrid for cultivation. Keeping in view following study was undertaken

Since 2009 every year around 50 most promising *Bt* cotton hybrids of different seed companies along with non *Bt* varieties and hybrids released from CCS Haryana agricultural University were planted and evaluated for seed cotton yield, earliness, insect –pest and disease data along with non *Bt* hybrids and varieties. The performances of these were sent to Department of Agriculture, Government of Haryana for their recommendation of most promising and de notification of least performing hybrids were discussed. Status of different *Bt* cotton hybrids in different years with respect to seed cotton yield were described along with their fibre quality and reaction to biotic stresses.

Seed cotton yield Two pickings were done in this experiment. First picking was done on 21.10.2012 i.e. 155 days after sowing and second picking on 19.11.2012 i.e. 184 days after sowing. On the basis of total kapas picked after second picking, the highest yield was recorded in KSCH 204 BG II (3340 kg/ha) followed by KSCH 211 BG II (3205 kg/ha). The lowest yield was recorded in the *Bt* hybrid PRCH711 BG II (58 kg/ha).

Boll weight: Maximum boll weight was recorded in Ankur 3228 BG II (4.96 g) followed by NBC 51 BG II (4.83 g). The lowest boll weight

Table 1. Performance of *Bt* cotton hybrids during 2010 and 2011 tested at CCS HAU, Hisar

Name of the hybrid/ variety	Seed cotton yield (kg/ha)			Ranking
	2010	2011	Average	
Pancham BG II	3436	3215	3326	1
Ankur 3028 BG II	3560	3009	3284	2
JK 1050 (JK Seeds)	3416	3138	3277	3
VICH 310 (Vikram Seeds)	3580	2803	3192	4
MRC 7017 (MAHYCO Seeds)	2922	3318	3120	5
MRC 7361 (MAHYCO Seeds)	2695	3524	3110	6
H 1226 Non *Bt* (Check variety)	3148	2855	3002	7
VICH 308 (Vikram Seeds)	2695	3 215	2955	9
RCH 605 BG II (Rasi Seeds)	2531	3241	2886	10
RCH 134 BG I (Rasi Seeds)	2263	2495	2379	Check
RCH 134 BG II (Rasi Seeds)	2963	2803	2883	Check
Bio 6488 BG I (Bio Seeds)	3086	2341	2714	Check
Bio 6488 BG II (Bio Seeds)	2963	2469	2716	Check

among the non *Bt* variety was recorded H 1226 (3.02g)

Ginning outturn : The ginning out turn ranged from 27.5 per cent (Ankur Jai BG II) to 35.3 per cent (Vardan BG II). In general *Bt* hybrids have less ginning out turn.

Fiber quality: The fiber of the all the *Bt* Cotton hybrids was sent for testing to CIRCOT (ICAR) laboratory for the testing of 2.5 per cent span length, Uniformity ratio, Micronaire value for fineness of the fiber and fiber strength.

2.5 per cent span length: The maximum 2.5 per cent span length was recorded in the hybrid Ankur Jai BG II (34.7mm) followed by Ankur 3228 BG II (34.1mm), NCS 459 BG II (33.4 mm), Western Nirogi 151 BG II (33.3mm). The minimum was observed in the non *Bt* check variety H 1226 (25.5mm) however among the *Bt* hybrids the lowest was recorded in the hybrids SP 7007 BGII and RCH 653 BG II 27 mm each.

Uniformity ratio: It was maximum in the *Bt* hybrids Kuber BG II (54%) followed by NBC 51 BG II and Vardan BG II (53% each). Whereas, it was lowest in the *Bt* hybrid MRC 7017 BG II (44%).

Micronaire value: Fineness of the fiber is measured by the micronaire value, lower the value, finer is the fiber and vice-versa. The lowest micronaire value was recorded in the hybrid Shakti 9 BG II (3.5) and highest micronaire value (4.9) was recorded in hybrids KSCH 204 BG II and RCH 569 BG II.

Fiber strength: Maximum fiber strength (tenacity) was observed in the hybrid Bioseed 6539 BG II (27.2 g/tex) whereas, lowest was recorded in the hybrid RCH 653 BG II (21.3 g/ tex).

Cotton leaf curl virus disease: Onlt two hybrids RCH 650 and Bio 6317 were free from cotton leaf curl virus disease.

Conclusion : On the basis of both the pickings i.e. first 155 DAS and second 184 DAS the hybrids KSCH 204 BG II, KSCH 211 BG II, RCH 653 BG II, Grand BG II, Border 507 BGII,

Mist BG II and KSCH 210 BGII were found promising as they yielded significantly higher than the popular *Bt* hybrids RCH 134 BG II, Bioseed 6488 BG II and Ankur 3028 BG II. The hybrids DPC 3083 BG II and Platinum 605 BG II yielded significantly higher than the popular *Bt* Hybrids RCH 134 BG II and Bio 6488 BG II only. The hybrids KSCH 204 BG II and KSCH 211 BG II yielded significantly higher than all the non *Bt* checks. The hybrids KSCH 204 BG II, KSCH 211, BG II RCH 653 BG II, Grand BG II and Border 507 BGII yielded significantly higher than all the non *Bt* checks except H 1098-i. The hybrid Mist BG II yielded significantly higher than the non *Bt* checks H 1117, H 1226 and H 1236.

Sr. No.	Name	Yield (kg/ha)
1	KSCH 204 BGII	3340
2	KSCH 211 BGII	3205
3	RCH 653 BGII	3162
4	Grand BGII	3162
5	Border 507 BGII	3087
6	Mist BGII	2967
7	KSCH 210 BGII	2840
8	DPC 3083 BGII	2792
9	Platinum 605 BGII	2777
RCH 134 BG II		1524
Bioseed 6488 BG II		2029
Ankur 3028 BG II		2321
Non *Bt* Variety check H 1117		1510
Non *Bt* Variety check H 1226		2478
Non *Bt* Variety check H 1236		2307
Non *Bt* Variety check H 1098-i		2692
Non *Bt* Hybrid check HHH 223		2549
C.D. (kg/ha) / C.V. (%)		**481/13.02**

Recommendation: It can be concluded on the basis of this year data that amongst all the *Bt* hybrids tested, the hybrids *viz.* KSCH 204 BGII, KSCH 211 BGII, RCH 653 BGII, Grand BGII, Border 507 BGII, Mist BGII, KSCH 210 BGII, DPC 3083 BGII and Platinum 605 BGII were found promising as they yielded higher than the popular *Bt* hybrids and non *Bt* checks.

Conclusion on the basis of three years data:

Name	Yield (kg/ha)			
	2010	2011	2012	Average
Ankur 3028 BG II	3560	3009	2321	2963
JKCH 1050 BG II	3416	3138	2293	2949
MRC 7017 BG II	2922	3318	2563	2934
Mist BG II	2263	3472	2967	2901
Pancham 541 BG II	3436	3215	1952	2868
H1226 (non-*Bt* check)	3148	2855	2478	2827
VICH 310 BG II	3580	2803	2087	2823
Bio 6488 BG II	2963	2469	2029	2487
Bio 6588 BG II	2593	3035	1759	2462
Bio 2113 Bunty BG II	2119	3138	2108	2455
RCH 134 BG II	2963	2803	1524	2430
NCS 855 BG II Raghav	2428	2418	2108	2318
MRC 7361 BG II	2695	3524	187	2135
Ankur Jai BG II	1955	2186	1980	2040
RCH 569 BG II	2942	2135	897	1991
VICH 309 BG II	2160	2135	1610	1968
Bioseed 6317 BG II	1955	2135	1638	1909
NCEH 31 BG II Yuvraj	1523	1852	557	1311

Recommendation : On the basis of three years data the *Bt* hybrids *viz.,* Ankur 3028 BG II, JKCH 1050 BG II, MRC 7017 (Nikki), Mist BG II, Pancham BG II, VICH 310 BG II, Bio 6488 BG II, Bio 6588 BG II, Bio 2113-2 (Bunty), RCH 134 BG II, NCS 855 (Raghav), MRC 7361 BG II and Ankur Jai BG II can be recommended for cultivation.

The hybrids RCH 773 BGII, RCH 653 BGII, Border, NCS 459 (Suma), Surpass IT 905, JK 0109 BGII, JK 1947 X – Gene, SO 7 H 878 BGII, MRC 7361 BGII, Bunty, RCH 650, PRCH 7799 (Zordar), RCH 776, KSCH 211 and SP 7010 BG II yielded more than27 q/ha and hence these hybrids can be recommended for cultivation.

On the basis of performance for seed cotton yield, maturity duration, insect pest and disease reaction and fibre quality data promising *Bt* cotton hybrids, non *Bt* varieties/hybrids were identified and recommendation is sent every year to Department of Agriculture, Government

Yield performance and reaction to biotic stresses of different hybrids/ varieties during 2013

S.No.	Variety/hybrid	Seed cotton yield (kg/ha)	CLCuD reaction	Whitefly/ leaf	Leaf hopper/ leaf	Thrips/ leaf	Boll damage by BW (%)	Locule damage by BW (%)
1	RCH 773	3549	MS	14.2	2.1	0.40	0	0
2	RCH 653	3025	S	16.5	0.5	0.40	0	0
3	Border	2994	S	19.3	0.4	0.27	2.96	0
4	NCS 459 (Suma)	2978	S	17.2	0.9	0.63	2.15	0
5	Surpass IT 905 *Bt*	2948	S	18.0	0.4	0.67	0	0
6	JK 0109	2917	S	15.9	0.6	0.07	0	0
7	JK 1947 X - Gene	2870	MS	20.1	0.5	0.67	0	0
8	SO 7 H 878 BGII	2824	MR	20.3	0.8	0.27	0	0
9	MRC 7361 BGII	2809	S	18.4	0.7	0.57	1.39	0.73
10	Bunty	2809	R	17.9	0.7	0.80	0	0
11	RCH 650	2747	MS	16.8	0.7	0.67	0.99	0.12
12	PRCH 7799 (Zordar)	2747	MS	16.5	0.5	0.53	0	0
13	RCH 776	2716	S	16.7	1.0	0.37	0	0
14	KSCH 211	2701	S	18.0	0.4	0.60	0	0
15	SP 7010 BG II	2701	S	19.7	0.5	0.47	0	0
16	KSCH 218	2562	S	13.8	0.5	0.40	0	0
17	MH 5302	2546	S	18.9	0.8	0.47	0	0
18	Bio 6488 BGII	2515	S	20.0	0.9	0.33	0	0
19	KCH 999 BG II	2469	HS	11.0	0.4	0.53	0	0
20	JK 1050 X - Gene	2469	S	19.0	0.6	0.47	0.9	0.12
21	Bio 6588 BG II	2438	S	16.2	1.2	0.40	0	0
22	RCH 791	2423	R	17.7	0.7	0.60	2.26	0
23	PCH 877 Leo cot BGII	2407	S	12.2	0.7	0.53	0	0
24	NCS 855 (Raghav)	2392	S	17.6	0.7	0.43	0	0
25	NCS 4455	2377	S	17.7	0.7	0.47	3.33	0
26	DPC 3083	2330	S	16.0	0.5	0.20	0	0
27	KSCH 213	2299	S	16.0	0.5	0.60	0	0
28	MRC 7041 BGII	2269	S	17.3	0.7	0.53	0	0
29	H 1226	2222	S	16.0	1.7	0.50	3.24	1.89
30	SP 7007	2222	S	12.8	0.7	0.53	0.79	0.21
31	HHH 223	2191	MS	13.9	1.6	0.43	1.1	0
32	MH 5304	2083	S	13.3	0.7	0.50	1.69	0
33	RCH 602	2068	MS	5.7	0.2	0.57	0	0
34	MRC 7017 BGII	2068	S	16.3	0.6	0.53	1.31	0.91
35	H 1300	2052	HS	15.2	1.3	0.40	0	0
36	H 1098-i	2037	S	17.2	2.0	0.73	2.33	1
37	Pancham 541	1975	HS	18.2	0.4	0.27	0	0
38	PRCH 333 Mahi BG II	1975	HS	15.9	0.7	0.47	0	0
39	Bio 6317	1944	S	17.7	0.5	0.63	0	0
40	SWCH 4713	1867	S	13.6	2.1	0.40	0	0
41	H 1236	1852	HS	16.6	2.0	0.43	2.06	0
42	VICH 310	1806	HS	19.3	0.5	0.47	1.09	0.13
43	JK 1050 BGII	1759	MS	17.9	0.4	0.47	0	0

Contd...

44	PCH 9609 Sirhind	1728	S	13.3	0.5	0.60	0	0
45	H 1117	1636	HS	16.7	1.9	0.63	1.64	0.82
46	RCH 134 BG II	1605	HS	18.0	2.1	0.40	1.29	0.16
47	KSCH 209	1590	HS	17.4	0.7	0.53	0	0
48	KSCH 210	1543	HS	19.4	0.7	0.40	0	0
49	NCS 9002 (Balwan)	1528	S	16.2	0.7	0.57	0	0
50	SWCH 4704 (US 21BGII)	1512	S	18.7	1.7	0.53	2.44	0
51	KSCH 215	1481	MS	15.6	0.6	0.47	1.45	0.18
52	RCH 134 BG I	1404	S	14.8	1.7	0.60	0.99	0.25
53	KCH 311 BG II	1327	HS	17.0	0.6	0.40	0	0
54	PRCH 711	1296	HS	11.1	1.0	0.23	0	0
55	Jadoo	1219	S	16.3	0.6	0.50	0	0
56	Western Nirogi 151	1219	S	19.8	0.6	0.57	1.45	0
57	KSCH 201	386	HS	15.9	0.6	0.53	1.56	0.79

0-10: Immune/ Disease free, 10.1-20: Resistant, 21-30: Moderately resistant, 31-40: Moderately susceptible, 41-50: Susceptible, Above 50: Highly susceptible, As per AICCIP CLCuD scale 0 -6 basis

On the basis of two years data i.e. 2012 and 2013 the following hybrids were found promising for cultivation in the Haryana state.

Name	Yield (kg/ha)		
	2012	2013	**Average**
RCH 653 BG II (Rasi Seeds)	3162	3025	**3093**
MRC 7017 BG II (Mahyco Seeds)	2563	2068	**2315**
RCH 650 BG II (Rasi Seeds)	2336	2747	**2541**
KSCH 201 BG II (Kohinoor Seeds)	2023	694	**1359**
SP 7007 BGII (Surpass)	1838	2948	**2393**
JKCH 1050 BG II (JK Seeds)	2293	1759	**2026**
Bio 2113 Bunty BG II	2108	2809	**2458**
KSCH 211 BG II (Kohinoor Seeds)	3205	2701	**2953**
Pancham 541 BG II (Krishidhan)	1952	1975	**1964**
H1226(non-*Bt* check)	2478	2222	**2350**
KSCH 218 BG II (Kohinoor Seeds)	2564	2562	**2563**
VICH 310 BG II (Vikram Seeds)	2087	1806	**1946**
Bioseed 6588 BG II	1759	2438	**2099**
NCS 855 BG II Raghav (Nuziveedu)	2108	2392	**2250**
RCH 134 BG II (Rasi Seeds)	1524	1605	**1564**
KSCH 209 BG II (Kohinoor Seeds)	2562	1590	**2076**
Bioseed 6488 BG II	2029	2515	**2272**
KSCH 210 BG II (Kohinoor Seeds)	2840	1543	**2192**
RCH 602 BG II (Rasi Seeds)	1268	2068	**1668**
NCS9002 BG II Balwan(Nuziveedu)	755	1528	**1141**
Bioseed 6317 BG II	1638	1944	**1791**
MRC 7361 BG II (Mahyco Seeds)	187	2809	**1498**
KSCH 213 BG II (Kohinoor Seeds)	2265	2299	**2282**

of Haryana so that it can encourage good performing hybrids/varieties and ban / discourage poor yielding hybrids to enhance the state cotton productivity along with other

management practices.

List of poor yielding *Bt* cotton hybrids on the basis their performance of 2013-14

Sr. No.	Name of hybrid/variety	Yield (kg/ha)	CLCuD reaction
1	KSCH 215	1481	S
2	RCH 134 BG I	1404	S
3	KCH 311 BG II (ATM)	1327	HS
4	PRCH 711 (Suraksha)	1296	HS
5	Jadoo KCH 14 K 59 BG	1219	S
6	Western Nirogi 151	1219	S
7	KSCH 201	386	HS

Recommendation: On the basis of four years data the *Bt* hybrids *viz.* MRC 7017 BG II, JKCH 1050 BG II, Pancham 541 BG II , VICH 310 BG II, Bio 2113 Bunty BG II, Bioseed 6488, Bioseed 6588 BG II, NCS 855 BG II Raghav can be recommended for cultivation.

Evaluation of *Bt* hyrids for restricted water availability : As water is becoming a limiting factor so it is also essential to identify the *Bt* cotton hybrids which perform better under restricted moisture condition. In this experiment irrigation was applied for sowing purpose only and after that crop was kept rain fed through the crop season.This experiment was done for the 2012 and 2013. The *Bt* cotton hybrid wewe selected which performed good under water stress conditions. Same hybrids also evalution

On the basis of four years data *i.e.* 2010, 2011, 2012 and 2013 the following hybrids were found promising for cultivation in the Haryana state.

Name	Yield (kg/ha)				
	2010	2011	2012	2013	**Average**
MRC 7017 BG II (Mahyco Seeds)	2922	3318	2563	2068	**2718**
H1226(non-*Bt* check)	3148	2855	2478	2222	**2676**
JKCH 1050 BG II (JK Seeds)	3416	3138	2293	1759	**2652**
Pancham 541 BG II (Krishidhan)	3436	3215	1952	1975	**2645**
VICH 310 BG II (Vikram Seeds)	3580	2803	2087	1806	**2569**
Bio 2113 Bunty BG II	2119	3138	2108	2809	**2543**
Bioseed 6488 BG II	2963	2469	2029	2515	**2494**
Bioseed 6588 BG II	2593	3035	1759	2438	**2456**
NCS 855 BG II Raghav (Nuziveedu)	2428	2418	2108	2392	**2336**
MRC 7361 BG II (Mahyco Seeds)	2695	3524	187	2809	**2304**
RCH 134 BG II (Rasi Seeds)	2963	2803	1524	1605	**2224**
Bioseed 6317 BG II	1955	2135	1638	1944	**1918**

Performance of *Bt* hybrids under irrigated and restricted irrigation (RI) conditions

Sr No.	Name of the *Bt* hybrid	2012Yield (kg/ha)		2013 Yield (kg/ha)		**Average yield(kg/ha)**	
		Irrigated	Restri. Irri.	Irrigated	Restri. Irri.	**Irrigated**	**Restri. Irri.**
1	DPC 3083	2792	3512	2330	1999	**2561**	**2756**
2	BUNTY	2108	3219	2809	2443	**2458.5**	**2831**
3	MRC 7017 (Nikki)	2563	3005	2068	2156	**2315.5**	**2581**
4	BIO 6588	1759	2222	2438	2770	**2098.5**	**2496**
5	SP 7007	1838	2606	2222	1228	**2030**	**1917**
6	PANCHAM 541	1952	2423	1975	1267	**1963.5**	**1845**
7	VICH 310	2087	2707	1806	879	**1946.5**	**1793**
8	RCH 134	1524	1856	2038	1254	**1781**	**1605**
9	MRC 7361	187	1418	2809	1790	**1498**	**1604**

under irrigated condition.

On the basis of mean performance both the years it can be conluded thath*Bt* cotton hybrids namely DPC 3083, Bunty,MRC 7017 and Bio 6588 performed better under restricted irrigation conditions as compared to irrigated condition. Hence their cultivation was advocated for areas with less water availability.

The seed development in some of the hybrids / varieties was very poor due to abnormal weather conditions and high incidence of sucking pests which resulted in excessively high ginning out turn and such inflated values may be considered as an exception case due to environmental factors.

Reaction of cotton leaf curl virus disease : The intensity of cotton leaf curl disease (CLCuD) reaction was recorded on 0 to 6 grades and per cent disease index was calculated as per standard AICCIP formula. The observation with regard to Cotton Leaf curl virus disease is given in below:

Highly resistant: 1 Bunty (Bio 2113-2)
Resistant hybrids: 2 (RCH 602 and RCH 650)

Moderately resistant hybrids: 5(H 1098-I, RCH 791, Super 6588, Bio 6588, RCH 653 and Ankur3224)

Boll weight: Maximum boll weight was recorded in SO 7 H878 BGII (4.00 g) followed by RCH 773 BG II (3.90 g). The lowest boll weight among the non *Bt* variety was recorded H 1226 and KDCHHN 9632 BG II (2.70 g)

Ginning Outturn: The ginning out turn ranged from 32.7 per cent (RCH 791 andPCH 9602) to 41.8 per cent (Bio 6165). In general ginning out turn during this year was on higher side due to poor development of seed.

Fiber quality: The fiber samples were sent to CIRCOT (ICAR) laboratory for the testing of 2.5 per cent Span length, Uniformity ratio, Micronaire value and Fiber strength2.5 per cent Span length:

The maximum 2.5 per cent span length was recorded in the hybrid Ankur 3244 (31.0 mm) followed by KCH 999 (30.3 mm), ATM and Bullet (29.5 mm). The minimum was observed in the non-*Bt* check variety H1117 (22.9 mm).

Uniformity ratio : Uniformity ratio determines the fiber quality by describing the uniformity in the fiber length. It was maximum in the *Bt* hybrids Bunty (52 %) followed by H 1117 (51%) and H 1226, KDCHH541, RCH 650, DPC 3085, RCH 773(50%). Whereas, it was lowest in the *Bt* hybrid PCH 9602 (44%).

Micronaire value: Fineness of the fiber is measured by the micronaire value, lowers the value, finer is the fiber and vice-versa. The lowest micronaire value was recorded in the hybrid KDCHH 516 and H1226 (3.8) and highest micronaire value (5.1) was recorded in hybrids Bio 6317-2 and DPC3083.

Fiber strength: Maximum fiber strength (tenacity) was observed in the *Bt* cotton hybrid Bullet (24.9 g/tex) whereas, lowest was recorded in the variety H 1117 (18.5 g/tex).

On the basis of seed cotton yield of both the pickings, the highest yield was recorded in *Bt* cotton hybrid RCH 602 BG II (2957 kg/ha) followed by ATM 311 (2530 kg/ha) and Bunty (2513 kg/ha). Other promising *Bt* cotton hybrids were RCH 791 BG II (2410 kg/ha), PCH 9604 (2325 kg/ha),SO7 H 878 BG II (2308 kg/ha), Super 6488 (2308 kg/ha), Bio 6165 (2291 kg/

Performance of varieties/hybrids evaluated during 2014 for different traits

Sr.No	Name	Mean Data Sucking pests population/leaf			Reaction to CLCuD	Seed cotton yield (kg/ha)	Per cent yield in first picking
		Whitefly	Leafhopper	Thrips			
1	RCH 602	8.00	0.00	2.17	R	**2957**	87.3
2	ATM (KCH 311)	12.33	0.25	0.00	S	2530	87.8
3	Bunty (Bio 2113-2)	10.50	0.25	0.42	HR	2513	94.6
4	RCH 791	11.75	0.83	0.92	MR	2410	93.6
5	PCH 9604	11.25	0.83	1.00	MS	2325	89.0
6	SO 7 H878 BGII	14.67	0.42	0.00	MS	2308	94.1
7	Super 6488 (6539-2)	13.33	0.50	0.00	MS	2308	90.4
8	Bio 6165	12.17	0.00	0.00	MS	2291	85.1
9	JK TARZAN	12.33	0.75	1.67	MS	2291	88.1
10	Ankur 3028	13.83	0.25	0.00	MS	2274	84.2
11	PCH 9609	10.25	0.33	0.83	MS	2239	85.5
12	Super 6588 (2510-2)	11.00	0.00	0.83	MR	2205	92.2
13	SP 7007	11.17	0.67	1.50	S	2188	90.6
14	NCS 9002 (Balwan)	14.33	0.17	2.42	MS	2188	88.3
15	Bio 6588	11.75	0.25	0.50	MR	2171	92.1
16	RCH 653	10.67	0.75	1.92	MR	2171	95.3
17	RCH 773	24.08	0.67	0.67	MS	2154	95.2
18	KCH 999	13.50	0.83	0.00	S	2154	67.5
19	NCS 855 (Raghav)	13.92	0.33	1.00	MS	2120	93.5
20	NCS 9013	12.00	0.33	2.17	MS	2120	92.7
21	RCH 650	10.92	0.17	1.17	R	2060	94.6
22	RCH 776	11.00	0.17	0.00	MS	2026	95.4
23	RCH 314	11.50	0.42	1.58	MS	2017	93.2
24	Bio 6488	13.08	0.50	0.00	MS	2000	85.5
25	JKCH 8940	13.67	0.00	0.00	MS	1966	94.8
26	H 1098-i	14.00	0.25	0.33	MR	1863	90.8
27	DPC 3085 BGII	11.92	0.42	1.08	MS	1863	95.4
28	Sikander	13.83	0.50	0.00	MS	1846	90.7
29	DPC 3083 BGII	16.33	0.58	1.25	S	1821	93.9
30	H 1226	11.42	0.17	0.67	MS	1778	88.5
31	PCH 877	18.67	0.17	1.50	MS	1761	91.3
32	HHH 223	10.67	0.42	0.67	MS	1726	94.1
33	Bio 6317-2	12.25	0.00	0.00	MS	1726	90.1
34	JKCH 1050	19.75	0.00	1.67	S	1726	**96.0**
35	SP 7010	10.75	0.75	0.50	S	1692	94.9
36	JKCH 0109 BGII	13.00	0.83	0.33	MS	1675	93.9
37	PCH 1414 (Leo cot)	19.17	0.42	1.83	S	1675	93.9
38	PCH 9602	20.17	0.67	2.58	S	1667	94.4
39	Ankur 3224	14.75	0.33	2.17	MR	1650	92.2
40	KDCHH 516	13.08	0.42	1.25	MS	1624	92.6
41	Ankur 3244	10.75	0.83	0.00	MS	1607	80.9
42	KDCHHN 9632	9.50	0.00	1.08	MS	1590	94.6
43	SP 7171	10.67	0.75	1.67	MS	1496	93.7
44	Bullet (KCH 707)	15.83	1.00	1.58	S	1470	80.2
45	JKCH 1947	13.83	0.25	2.50	MS	1436	92.9
46	H 1300	15.58	0.17	0.50	S	1350	92.4
47	H 1117	12.58	0.50	0.17	S	1333	84.6
48	KDCHH 541	18.33	0.33	2.25	S	1265	91.9
49	Jadoo (KCH 14K59)	15.58	0.50	1.33	S	1214	88.7
50	H 1236	16.58	0.83	0.00	S	1111	89.2
51	Jackpot	11.92	0.17	1.42	MS	923	83.3
	Range	8.0(RCH 602) - 24.08(RCH 773)	0.0-1.0	0.0-2.58	HR - S	2957(RCH 602) - 923(Jackpot)	67.5 (KCH 999)- 96.0 (JKCH 1050)

ha), JK Tarzan (2291 kg/ha), Ankur 3028 (2274 kg/ha), PCH 9609 (2239 kg/ha), Super 6588 (2205 kg/ha). These hybrids have good seed cotton yield and moderately susceptible or moderately resistant reaction to cotton leaf curl virus disease.

Only one *Bt* cotton hybrid RCH 602 BG II (2957 kg/ha) significantly out yielded the most popular check hybrid Bio 6588 BG II (2171 kg/ha). The lowest yield was recorded in the *Bt* hybrid Jackpot (923 kg/ha).

Cotton wheat relay cropping : This experiment was started in the year 2013 and it was repeated in 2014. During the year 2013 a total of four pickings were done. First picking was done on 18.1.2013, second on 15.11.2013 as conventional practice and after wheat crop was sown in these plots and third picking on 17.12.2013 and forth picking was done on 18.01.2014. Under relay cropping system a yield from 15 kg/ha to 336 kg/ha were harvested in wheat crop.

During 2014 under relay system seed cotton yield was less than 50 kg/ha in third picking and no need to go for fourth picking. From this experiment it was concluded that under Haryana condition relay cropping of wheat is not recommended if normal maturity duration hybrids/varieties are planted because they produce better seed cotton yield under conventional practice i.e. about 180 days after sowing than late maturing hybrids (normal + relay yield). This practice may be recommended for specific conditions like due to some environmental conditions crop maturity period may be prolonged further and secondly a farmer had grown late maturity duration hybrid by unknowingly.

third picking and in fourth picking there was no yield from any of the plot from different BT hybrids and non *Bt* varieties/hybrids ranged

Pest status:

A: Observation at the time of sowing wheat crop: 18:11:2013

1. Bollworm infestation was not found in any of the entry.
2. Sucking pests (thrips, leafhopper, aphid and whitefly) infestation was also not observed.

B: observation on 9:1:2014

1. In non *Bt* cotton hybrids bollworms infestation was observed from 0.60 to 1.55 per cent but bollworm infestation was nil in *Bt* hybrids.
2. Aphid infestation was observed in cotton flower buds (10-15 aphids) but not on

S. No.	Entry code	Per cent bollworm infestation	Aphid/flower bud
1	H1117 (non *Bt* check variety)	0.80	18
2	H1226 (non *Bt* check variety)	1.55	15
3	H 1236 (non *Bt* check variety)	0.60	20
4	H1098-i (non *Bt* check variety)	1.39	12
5	Bio 6488 BGII	0.00	10
6	Bio 6588 BG II	0.00	15
7	Jadoo KCH 14 K 59 BG	0.00	10
8	JK 1050 BG II	0.00	13

Characterization of work related drudgery of women in cotton production

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Abstract : Considerable research has been done by All India Coordinated Research Project on Home Science, to understand the work and associated drudgery of women in specific production activities, their access to technology and its implications on gender health and productivity. The key factors of human labor interface in activities viz; time load, physical load, postural load, repetitive strain, physiological load, musculoskeletal load were examined as dimensions to assess the work related drudgery load pertaining to cotton production system. The present paper is based on the data pertaining to Telangana collected from 30 women farmers involved in cotton production. The specific findings include that in Telangana , women exclusively participated in removing stalks, dibbling, weeding and crop harvesting. Land tillage, row marking, interculture operations, pest management were considered as men exclusive tasks. Majority of the activities done by women were being manually performed. As per work load, Removing stalks and stubbles, spreading of manure and harvesting were categorized as having high level work load. Whereas sowing , weeding were found to be having very high level load leading to drudgery. Statistically, significant variation was confirmed between factors of drudgery dimensions were considered. As per test of associations, drudgery in the activity removing of stalks and stubbles was found associated with posture, spreading of manure was associated with posture, physiological load and MSD (Musculo Skeletal Disorder). Sowing activity was associated with Posture, Repetitive strain and Time load and weeding activity was associated with physiological load. Drudgery in harvesting activity was associated with physiological load, Repetitive strain, Time load and MSD at 5% level. The results lead to technology interventions in the production system to mitigate drudgery.

Key words : Characterization of drudgery, cotton production, farm women

Cotton plays a vital role in Indian agricultural economy and offers employment for about 60 million people. Telangana is one of the major cotton-producing states of India. The industrial production of cotton seed is also concentrated in the state. Cotton is grown as the main crop in Warangal and Mahabubnagar. Telangana farmers grow cotton in 14 lakh hectares during a normal *kharif* season under rain fed conditions. Women workers in majority are preferred as workers in commercial agriculture like tea, coffee, sugarcane, cotton, tobacco and plantation products (Singh *et al.,* 2007). Dibbling, weeding and hand picking are the important productions related activities that demand women labour. In recent years, labour shortages during peak periods of cotton production; have been quite frequent and widespread. Traditionally, women do the exclusively tedious, time and labour intensive works resulting in fatigue and drudgery (Shilparani, 2007). To relieve the drudgery of women in production system activities, it is needed that the activities are characterized by associated factors contributing to drudgery so that suitable interventions could well be designed. Therefore, the study was planned with an objective to characterize drudgery by the

factors most associated with it.

REVIEW OF LITERATURE

Chayal and Dhaka (2010) analyzed the work participation of women in agriculture in Bundi district of Rajasthan. A total of 200 farm women selected as respondents through proportionate random sampling. The selected respondents were interviewed personally using pre-tested well structured interview schedule. The findings showed that farm women's participation was maximum in cutting, picking, cleaning of grains, drying of grains, storage, processing, weeding, winnowing and major part of cleaning of field, raising. Participation of farm women in agriculture was significantly affected by socio-economic variables like –age, family income, land holding.

In 2009, the Centre for Development and Environment (CDE), University of Bern, carried out an impact study to assess the economic, social and environmental impacts of organic cotton production in Jalalabad Oblast. The study reported that Organic farming increased the workload as more manual work was found necessary for applying manure, weeding, etc. Manual work was generally women's work. Thus, women more strongly perceived increase in workload since conversion to organic farming. This development was further aggravated by widespread male labour migration which resulted in more work and responsibility for women, both for organic and conventional farmers.

A study was engendered by the need to document the serious human health consequences of the indiscriminate use of pesticides on cotton in India by Mancini *et al.,* (2005). Fifty women cotton farmers from three of the villages located in Warangal and Mahabubnagar districts of Telangana formed as sample for the study. Results highlighted that the typically female tasks such as mixing concentrated chemicals and refilling spraying tanks were found as hazardous as direct pesticide application. Of 323reported events, 83.6 per cent were associated with signs and symptoms of mild to severe poisoning, and 10% of the pesticide application sessions were associated with three or more neurotoxic/ systemic signs and symptoms typical of poisoning by organophosphates, which were used in 47 per cent of the applications. Although in 6 per cent of the spray sessions the workers' neurotoxic effects were extremely serious, none sought medical care. Low-income marginal farmers were found more often subjected to severe poisoning than were landlords.

Sunita *et al.,* (2012) conducted a study on drudgery reduction of farm women with cotton picking bags. Picking efficiency, energy expenditure, carrying capacity, ease, comfort, safety, loading and unloading etc. were evaluated between Hisar and Prabhani designed cotton bags. Results revealed that cotton bags designed by Hisar was having 50 per cent higher carrying capacity, ease in tying, 37 per cent less load on heart beats, 18% lower energy (kJ)expenditure and proved significantly superior over Parbhani designed picking bags. Hisar bag required 25% and 15 per cent extra cloth and costs respectively over Parbhani bag. No significant difference was reported due to age and type of bag.

A study by AICRPH (2004) and DDK (2007) observed that cotton picking manually involved a lot of drudgery due to posture and abrasion of fingers due to sharp points of dried bracts. Through the efforts of testing and popularizing cotton harvest bags, they opined that picking

efficiency was increased and labour costs, trash contents were decreased.

Narinderjit *et al.,* (2007) purposively selected 60 female respondents who were intensively involved in cotton-picking activity in Bathinda district of Punjab state. Field experiments were conducted to compare the ergonomic cost in terms of physiological responses between conventional and improved techniques of cotton picking (improved bag and plucker). Results of the study exhibited significant reduction in Heart Rate (7.29%), Energy Expenditure (17.30%), Total Cardiac Cost of Work (43.75%) and Physiological Cost of Work (43.76%) with the use of improved methods. Women adopted improved bag and plucker and as users, they were satisfied and relieved of their drudgery.

METHODOLOGY

Characterization in the context is the concept of portraying the qualities of an activity in the selected production system that is either constraining the effective work performance of a worker or causing risk to health and safety of worker. Bench mark survey method was followed based on criteria of accessibility, willing cooperation of respondents, five villages were selected and made operational for the present study. They were drawn from two mandals namely, Moinabad and Chevella of RR district from Telangana state. Kethiradipalli, Tolkata, Bakaram, Ethabarpalli and Nagireddyguda therefore were formed as operational villages as cotton growers were found to an extent of 300 from marginal landholdings.

The survey was planned after all the production related activities were identified and were made into sub tasks. Among them, only the women exclusive and women dominant or women equal participation tasks were selected for characterizing activities as per factors during survey. Thirty women farmers representing 10 per cent of cotton growers from the operational villages were selected for the survey. Interview schedule was developed and standardized to collect the data from farm women by recall method. The interview schedule contained general information, where in details on subject's age, years of farming, family size and income, land holding status, crop calendar, gender participation and technology used were elicited apart from six variables viz; physical load, posture load, repetitive strain load, physiological load, time load and musculoskeletal disorder load. Each factor was measured using quantitative and qualitative methods as furnished in Table 1.

Activities were considered as independent and the six factors as dependent variables and a null hypothesis was formulated for the purpose of understanding the source of variation from among the activities and factors for the purpose of the study.

N_0: There is no significant variation in drudgery load between activities.

There is no significant variation in drudgery load due to factors.

Analysis of variance and chi square tests were conducted to confirm and characterize drudgery. Total drudgery was calculated using linear combination method as per the formula given below.

Total drudgery = (dr(PL) + dr(P) + dr(RS) + dr(T) + dr(MSDs) + dr(PysL)

Where; dr (total)= Total drudgery ;

PL – physical load (25 points) ;

P – postural load (25 points) ;

RS – repetitive strain load (25 points) ;

T – time load (25 points) ;

MSDs – musculoskeletal disorders (25 points) ;

PhsL – physiological load (25 points)
Drudgery Index %(DI) = [100 * dr (total)] / 150

Drudgery level categorization was done as follows

Assuming that manual physical works done beyond one third (30%) human capacity may be treated as heavy, the drudgery index was categorized for interpretation as below. It also equated to the physiological load calculated by heart rate method (Table 2)

Table 2. Drudgery Index categorization

Drudgery Index	Expected equivalent heart rate
<10(%) = Very low	
10–20(%) = Low	Up to 90 b/min
20–30(%) = Moderate	91 - 105 b/min
30–40(%) = High	106 - 120 b/min
40–50(%) = Very High	121 - 135 b/min
>50(%) = Extremely High	136 - 150 b/min

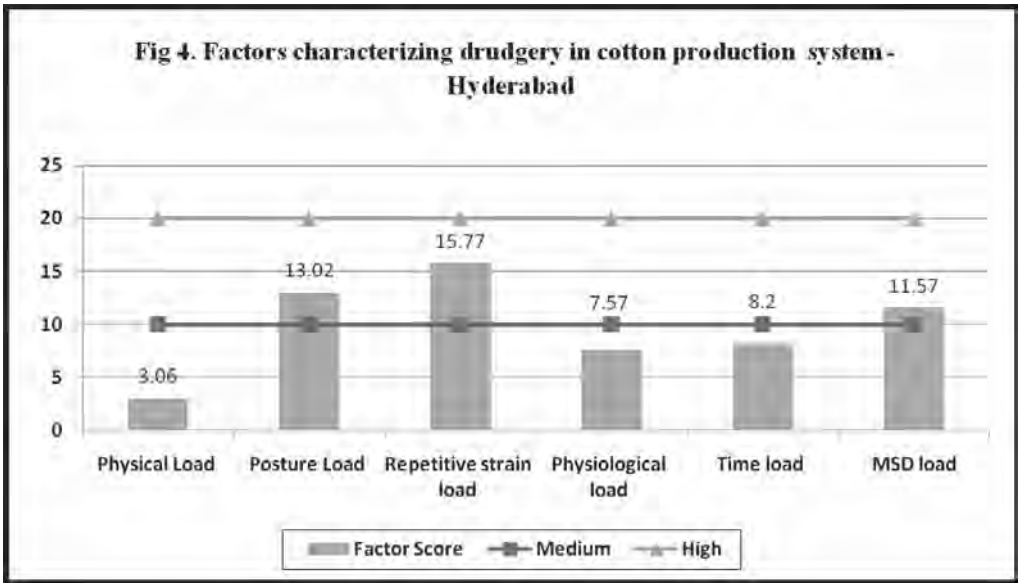
Table 1. Drudgery factors and their measurement

S. no.	Variables	Attributes
1	Physical load	a. Weight of the load (kgs)
		b. Distance carried (kms)
		c. Height lifted (mts)
		d. Physical load rating(5 point scale)
		e. Physical load factor
2	Posture	a. Nature of posture
		b. Body part involved
		c. Discomfort rating (5 point scale)
		d. Posture load factor
3	Repetitive strain	a. Nature of work
		b. Repetitive strain rating (5 point scale)
		c. Repetitive strain load factor
4	Physiological load	a. Physiological load r ating (5 point scale)
		b. Physiological load factor
5	Duration / time	a. Hours / day
		b. No. of days
		c. No of labour employed
		d. work load as per time
		e. Time load factor
6	Body pain and disorder	a. Body part involved
		b. Body disorder symptoms
		c. Body pain rating (5 point scale)
		d. Frequency
		e. Posture load factor

RESULTS AND DISCUSSION

Crop calendar : Cotton production activities were being done for seven months during Kharif in Telangana region. Mostly it was grown under rain fed conditions as commercial crop. Land preparation activities namely tillage, removing stalks and stubbles took place in the month of June and July with the onset of monsoon. Manuring and fertilizer application was done before sowings from mid july to August. Sowing was mostly done by dibbling method and after 15 days gap filling was done. Inter culture operations and weeding between plants was done during September and October and later for every 15 days. During this time, necessary pest management also was done. November and December were the months for cotton picking.

Gender participation : Gender participation in cotton production tasks revealed that , women exclusively participated in removing stalks, sowing, weeding and crop harvesting. Land tillage, row marking, interculture operations, pest management were considered as men exclusive tasks. Women belonging to Small and marginal farm category spent one or two days in their own farm followed by extending as wage labor for about 10 – 15 days in the village in other farms. Hand weeding was being done twice per season in addition to inter



culture operations. Inter culture operations were being done 6 times that means for every 10 to 15 days between rows. Cotton harvesting by manual picking method was being practiced and about three pickings were done as minimum if the crop did not fail.

Technology use : Majority of the activities done by women were being manually performed. Removing stalks was being done by manual pulling and gathering them for burning in the yard. Dibbling activity was being done after marking the rows and sometimes even between plants as 2ft and 1 ft respectively. Spreading manure was another activity done by women with the help of local baskets made of iron. Traditional kurpi was the common tool used for hand weeding and cotton harvest was done hrice in the season by manual picking using bear hands. Wooden plough was used for marking rows and tillage was being done by using tractor and intercultural operations with local cattle drawn hoe (g*untuk*a)

Drudgery load as per activity : As per drudgery index, viz., Removing stalks and stubbles ( ), spreading of manure ( ) and harvesting( ) were categorized as having high level drudgery where as sowing ( ), weeding ( ) were found to be having very high level drudgery. As per ANOVA, there was no significant variation between activities when drudgery load was considered. This infers that all the activities were dependent on the drudgery factors. Chi-square test of association was conducted for the sample based on major population distribution behavior.

As per test of associations, drudgery load while removing of stalks and stubbles was found highly associated with posture, spreading of manure was associated with posture, physiological load and MSD, sowing activity was associated with posture, repetitive strain and Time load, weeding activity was associated with physiological load and MSD and drudgery load in harvesting activity was associated with physiological load, repetitive strain, Time load and MSD at 5 per cent level. The results lead to the carefully planning needed while conducting technology interventions in the production

Table 1. Drudgery load as per activity

Farm activity	Removing stalks and stubbles	Spreading of manure	Sowing - dibbling	Weeding - plant to plant	Harvesting	**Factor wise Drudgery load**
Physical Load	3	3	3	4	2	15
Posture Load	9	14	20	19	4	65
Repetitive strain load	20	8	15	20	16	79
Physiological load	3	16	3	8	8	38
Time load	6	3	12	4	16	41
MSD load	15	13	10	8	12	58
Activity wise drudgery load	**55**	**57**	**63**	**63**	**58**	
Drudgery Index	**37**	**38**	**42**	**42**	39	

Table ANOVA for drudgery load on cotton production activities and factors n=30

Source of Variation	SS	df	MS	F	P value	F critical
Factors	510.61	5.00	102.12	3.81*	0.01	2.71
Activities	8.39	4.00	2.10	0.08	0.99	2.87
Error	536.07	20.00	26.80			
Total	1055.07	29.00	1055.07			

*Significant at 5 per cent

system.

Factors contributing to drudgery : Among the factors impacting overall drudgery in cotton production system activities, it was indicated that repetitive strain load followed by posture and MSD load were contributing in a priority order. The test of ANOVA, confirmed the significant variation in drudgery load attributed to the drudgery factors. Posture had thirty three percent of variation in drudgery load due to sowing, weeding and manuring activities. Twenty three percent of variation in drudgery due to factors was from physiological factor and manuring was found to be involving physiological drudgery. Twenty percent of the variations contributed by time factor were found due to harvesting activity. Though physiological and

Activities	Physical Load	Posture Load	Repetitive Strain	Physiological Load	Time Load	MSD	Drudgery Load
Removing stalks and stubbles	0	15	0	0	0	8	30
Spreading of manure	2	30	0	30	0	21	30
Weeding - plant to plant	0	0	0	30	0	30	30
Sowing - dibbling	0	30	30	0	30	10	30
Harvesting - Picking	0	0	30	30	30	30	30

time loads were considerable to handle, they were not rated important as per priority order by farm women. Women while performing the pulling and gathering of small stalks, weeding, sowing handle approximately 3kg of weight at a time and walk up to 2 km distance in each of the activities. However, women did not perceive while rating physical loads as priority and that was probably the reason for the low factor load on account of physical loads. The discomfort rating arising due to postures on body parts were found to be more while dibbling, weeding and spreading manure. But drudgery on account of postures was rated as low in priority while removing stalks and harvesting and therefore posture was found to be contributing to moderate drudgery. Repetitive strain load Repetitive strain factor was observed to be contributing to a moderate extent to drudgery of women Working while removing stalks and weeding.

CONCLUSIONS

The review and empirical evidence from the data used in the paper leads to conclude that the work of women in cotton production needs to be understood from the perspective of its implications on labour time and health. The resultant drudgery load varied based on activities and factors from low to very high level. As per ANOVA, there was significant variation between activities when factors were considered. This infers that all the activities were dependent on the drudgery factors. The technology options needs to be popularized for mitigating drudgery.

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Water management technologies for cotton under irrigated conditions

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Irrigation in agriculture has been an effective way to cope with the ever increasing food and fiber demand of India. Increasing the efficiency of water use by various crops continues to be a topic of concern because of increasing demand for water use and improved environmental quality. In recent years water resources available for irrigation have been decreasing due to the growing demand in municipal and industrial water uses. Cotton (*Gossypium hirsutum* L.) is one of the most important fiber producing crop grown in India. In northern India, crop is cultivated under irrigated conditions particularly in that areas where majority of underground water is of poor quality and the only source of good quality water for irrigation is river water supplied through canals which is inadequate quantity. Therefore, it has become an important issue to improve crop water use efficiency through proper irrigation design and management. There is great need for judicious use of canal water because excess usage causes deep percolation and results in high water table and secondary salinization. By adopting specialized and efficient methods of irrigation, the twin objectives of higher productivity and rational use of water can be achieved. As the various water saving technologies developed not only sustain the crop productivity but also save considerable amount of irrigation water without any significant losses in crop yield. Some of the improved technologies viz. border design, border slope, planting cum Irrigation methods, alternate furrow irrigation for poor quality water, timing of first irrigation, drip irrigation and fertigation in cotton showed the promising results in increasing water use efficiency in cotton under north India conditions.

1. Width of the border and slope of the field : The width of border and slope of field plays significant role in water management. The border width of 15 m gave 7.5 and 3.8 per cent more irrigation application efficiency as compared to 10 and 20 m border width on sandy loam soil conditions . The water application efficiency was similar under 0 and 0.1 per cent slope and decreased to 66.9 per cent with further increase in slope to 0.2 per cent on sandy loam soil.

2. First irrigation to cotton: Application of first irrigation too early or too late reduces seed cotton yield. It was observed that delay of first irrigation up to some period helps in improving the seed cotton yield as compared with application of first irrigation early at 30 DAS. Buttar *et al* (2007) also reported that delay of first irrigation from 28 days after sowing (DAS) to 42 DAS, resulted in an increase of 8, 14 and 17 per cent in seed cotton yield during first, second and third year of study respectively (Table 1). Delayed first irrigation from 28 DAS to 42 DAS also resulted in higher water expense efficiency (Table 2)

The initiation of irrigation at 28 DAS restricted root growth to surface layers which confined to 0-60 cm soil depth throughout the

Table 1. Effect of timing of first irrigation on seed cotton yield (Kg/ha)

Timing of first irrigation	Seed cotton yield (kg/ha)
2000	
28 DAS	1417
35 DAS	1449
42 DAS	1529
2001	
28 DAS	1110
35 DAS	1121
42 DAS	1270
2002	
28 DAS	1825
35 DAS	1985
42 DAS	2132
LSD (0.05)	
2000	103.4
2001	80.7
2002	126.3

Buttar *et al.,* (2007)

Table 2. Water Expense efficiency (kg/ha/cm) as influenced by timing of first irrigation

Timing of first irrigation	Seed cotton yield (kg/ha)
2000	
28 DAS	22.0
35 DAS	23.8
42 DAS	27.7
2001	
28 DAS	14.0
35 DAS	14.1
42 DAS	16.1
2002	
28 DAS	18.9
35 DAS	20.3
42 DAS	21.2

Buttar *et al.,* (2007)

cropping season (Fig1). On the other hand, higher root mass density and vertical distribution up to 180 cm was observed when the initial irrigation

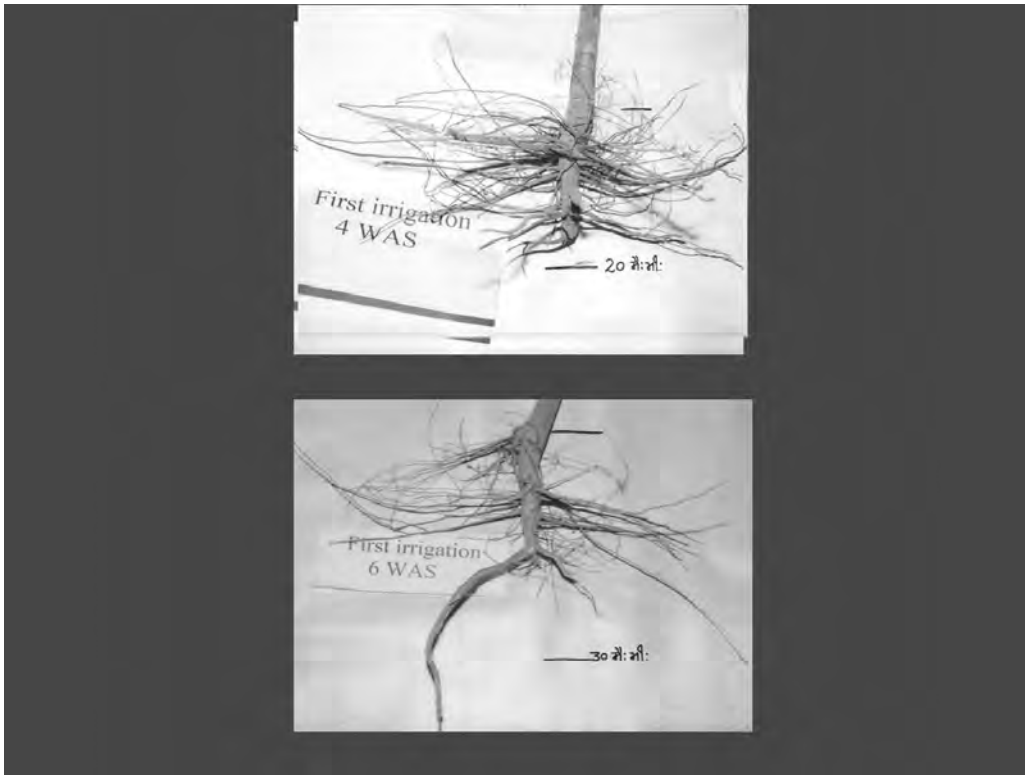

Fig. 1. Root growth under different irrigation schedules

was applied at 42 DAS. There was no vertical root growth where first irrigation was applied at 28 DAS (Buttar *et al.*, 2009).

3. Bed Sowing in cotton: The flood irrigation method of irrigation in cotton consume large volume of water, hence, there is need to switch over to more efficient systems of irrigation . Among these methods furrow irrigation is economical having little system cost. The planting of cotton on ridges with each furrow irrigation resulted in saving of 16.7 per cent irrigation water as compared to conventional method in *desi* cotton and 33.3 - 45.0 per cent saving in irrigation water in American cotton.The experiments conducted at Ludhiana and Bathinda showed that ridge sowing in cotton and irrigation application in furrows results in higher seed cotton yield coupled with higher water use efficiency (Table 3).

Table 3. Seed cotton yield and water use efficiency in cotton as affected by irrigation methods (Mean of 6 years)

Irrigation methods	Seed cotton yield (kg/ha)	Water use efficiency (kg/ha/cm)
Flood	1339	43.6
Each furrow	1390	51.7

Aujla *et al.,* (2001)

4. Alternate furrow irrigation for poor quality water : In Indo Gangetic plains, where the soils of cotton growing region are light in texture and under ground water is brackish, surface irrigation (check-basin) with poor quality waters usually lead to build up of salinity and sodicity problems and thus unsustainable cotton yields. Dwindling supplies of good quality water for irrigation and increasing demand from other users are forcing farmers to use saline/ sodic underground water for irrigation purpose.Surface irrigation (check basin) with poor quality waters usually leads to build up of salinity and sodicity problems and thus unsustainable cotton crop yields. It has been emphasized that when poor quality waters are used for irrigation due attention should be given to minimize root zone salinity. It has also been advocated that there is great need for selection and use of appropriate irrigation systems and practices that will supply just sufficient quantity of water to the root zone to meet the evaporative demand and minimize salt accumulation in the root zone . When using poor quality waters, the period of germination and emergence of the seedlings is the most critical stage of crop growth, and crops can tolerate higher salinity once good quality water was substituted for pre sowing irrigation to leach out the salts from the seeding zone. In Indo Gangetic plains where the soils of cotton growing region are light in texture and underground water is brackish, farmers grow cotton in rotation with wheat by applying four to five irrigations to each crop through flood irrigation method (check basin). In this region, the only source of good quality water for irrigation is river water supplied through canals.The sustainable use of underground poor quality water are the need of the hour for sustaining the land resources. Therefore, there is need to adopt specialized and efficient methods of irrigation that can help in attaining the twin objectives of higher productivity and rational use of poor quality water.

The results showed that in cotton, poor quality tube well water significantly reduced the seed cotton yield . The pre sowing irrigation with canal water and all subsequent irrigations with tube well water improved the seed cotton yield when compared with tube well water alone. However, this yield increase was significant only

in alternate furrow irrigation, and the yield obtained was at par with yield under alternate furrow in CW. When compared to check-basin irrigation, each furrow and alternate furrow irrigation resulted in a saving of 20 and 42 per cent respectively.

Table 4. Seed cotton yield (kg/ha) and water use efficiency (kg/ha/cm) as influenced by irrigation water quality and method of irrigation

Treatments	Seed cotton yield (kg/ha)	Water use efficiency (kg/ha/cm)
Canal water		
Check basin	1695	0.182
Each Furrow	1613	0.187
Alternate Furrow	1551	0.197
Tubewell water		
Check basin	1329	0.140
Each Furrow	1308	0.147
Alternate Furrow	1179	0.143
Canal water+tubewell		
Check basin	1453	0.154
Each Furrow	1384	0.159
Alternate Furrow	1463	0.184
LSD (0.05)	154	0.011

Thind *et al.,* (2010)

5. Drip irrigation and fertigation: In drip irrigation the volume of wetted soil at a particular water application is controlled by the volume of water added, the discharge rate of dripper and the soil water content . Drip method of irrigation is most suited to semi arid and arid areas where water is scarce and low water consuming and high value crops can be grown. The results showed that paired row planting (60 x 60 x 120 cm) increased seed cotton yield by 7.5 per cent over single row planting (60 x 90 cm) with higher water use efficiency. The application of 100 per cent recommended dose of N (150 kg N/ha) and K (20 kg K_2O/ha) through drip in six

Table 5. Soil pH and electrical conductivity as influenced by irrigation water quality and method of irrigation

Treatments	pH	Electrical Conductivity (dS/m)
Canal water		
Check basin	8.56	0.148
Each Furrow	8.51	0.139
Alternate Furrow	8.54	0.136
Tubewell water		
Check basin	8.99	0.302
Each Furrow	8.92	0.291
Alternate Furrow	8.94	0.289
Canal water+tubewell		
Check basin	8.94	0.274
Each Furrow	8.91	0.263
Alternate Furrow	8.89	0.256

Thind *et al.,* (2010)

equal splits at an interval of 15 days gave 49.8 % per cent higher seed cotton yield than control (flood irrigation).Drip irrigation under normal sowing (NS) resulted in an increase of 258 and 453 kg/ha seed cotton yield than check-basin during first and second year, respectively, when same quantity of water and N was applied. Drip irrigation under dense paired sowing (DP) in which the quantity of irrigation water applied was 75 per cent as compared with NS, further increased the yield by 84 and 101 kg/ha than NS during first and second year, respectively (Table 6) . Drip irrigation under NP, in which the quantity of water applied and number of laterals used were 50 per cent ascompared with drip under NS, resulted in a reduction in seed cotton yield of 257 and 112 kg/ha than NS during first and second year, respectively. However, the yield obtained in nprmal paired sowing (NP) under drip irrigation was equivalent to yield obtained in NS under check basin during first year but 341 kg/ha higher yield was obtained during second year. The decrease in N applied, irrespective of methods of planting, caused a

Table 6. Seed cotton yield as influenced by methods of planting, method of irrigation and nitrogen application

Methods of planting	Check basin	D_{100}	D_7	D_{50}	**Mean**
2003					
Nornal sowing	1656	1914	1585	1483	**1660**
Dense paired sowing (35x55x35 cm)	2084	1998	1553	1435	**1768**
Normal paired sowing (35x55x35cm)	1245	1657	1527	1235	**1416**
Mean	**1662**	**1856**	**1555**	**1384**	
LSD (0.05) Method of planting	159				
Rates of nitrogen	139				
Interaction	241				
2004					
Nornal sowing	742	1195	769	682	**847**
Dense paired sowing (35x55x35 cm)	1163	1296	1067	792	**1080**
Normal paired sowing (35x55x35cm)	636	1083	752	674	**736**
Mean	**847**	**1192**	**863**	**716**	
LSD (0.05) Method of planting	122				
Rates of nitrogen	97				
Interaction	168				

significant decline in seed cotton yield during both the years. Water use efficiency (WUE) under drip irrigation increased from1.648 to 1.847 and from 0.983 to 1.615 kg/ha/1 mm during first and second year, respectively, when the same quantity of N and water was applied. The WUE further increased to 2.125 and 1.788 kg/ha/mm under DP during first and second year, respectively.

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Mechanization of cotton cultivation in India

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Mechanization is a labour augmenting technology increasing output per worker and not directly contributing to yield as such. Benefits of mechanization are reduced drudgery, increased returns and reduced costs. Higher profit margins due to increased input use efficiency and increased yields due to timeliness of operation. Benefits of mechanization programme are greatest where labour is scarce and expensive as found during peak periods.

Cotton is cultivated in three distinct agro-ecological regions (North, Central and South) of the country. It is cultivated by 6.4 million farms, a quarter has less than 1 ha and half the farms are less than 2 ha in size. Of the total cotton growing area in the country, 65 per cent is rained (Rafiq and Pandolph, 2014). India produces a largenumber of cotton varieties and hybrids. The number of varieties in cultivation is more than 75, but 98 per cent of the production comes from 25 varieties (Osakwe, 2009).The soils of Central zone and some part of south zone contain large proportion of clay thus become very sticky when wet and very hard when dry. Sophisticated farm power systems are more suited to large land holdings of North zones coupled with higher power availability of around 3.5 kw/ha. In small and marginal farms, except for tillage, other operations such as sowing, weeding, cotton picking harvesting and stalk uprooting are normally manually performed. Though, India has abundant labour force in agriculture, non-availability of manpower during peak crop season is a growing problem.

A brief review of the status of cotton mechanization and research carried out in the cotton growing zones of India is presented in the following paragraphs.

Tillage equipment : Usage of tractor operated implements for tillage is fairly wide spread, especially in Haryana and Punjab. The soils of this region, sandy and sandy loams develop compacted layers within the soil profile that may impede proper root development of crop. Destruction of these compacted zones has been a focal point. Sub soiling, primary method of reducing soil compaction, has been successful in increasing cotton yields in most soils where compaction is a problem. The first dry cultivation is done with mould board/lister plough to enable deep cultivation. Cultivators using different types of shovels depending upon local conditions is quite popular with cotton farmers of north zone. It gives a tillage depth of 10-12 cm and prepares a clean, clear seed bed. Land levelers and graders are used for leveling alongwith plankers or *Pata* for land smoothing. Irrigation bunds are formed using disc type bund formers. In Punjab, irrigation and seedbed preparation were main energy consumers (Sodhi *et al.,* 1985). Improved equipment such as rotavator and rotary augur plough can be used to effect saving in Time, Energy and Cost, and enhance the quality of seedbed (Verma *et al.,* 1996).

In central cotton growing zone, the animal drawn wooden blade harrow is the most popular and often the only implement available

for secondary tillage operation in small and marginal farms (Plate 1).These implements have been improved by making them adjustable and replacing the traditional wooden structure with a common metallic frame and a set of different size blades with quick coupling and de-coupling arrangements. These are durable, lighter and cheaper.

Traditional Improved

Plate 1. Wooden bladeharrow

Table 1. List of tillage equipments used in India

Name of Equipment	Used for
Mould board plough	Dry cultivation
Lister plough	Dry cultivation
Disc harrow	
Cultivators	It gives a tillage depth and prepares a clean, clear seed bed
Land levelers	Leveling along with plankers for land smoothing.
Graders	Leveling along with plankers for land smoothing.
Blade harrows and hoes	Harrowing and hoeing

Planting equipment : For precision planting of cotton seed with machines it is necessary to make it free flowing by removing the fuzz around the seed by acid, known as acid delinting. This characteristic becomes more important as high speed planters are used. Delinted seed can be metered more accurately than fuzzy seed, which is highly desirable when planting to a stand.

Precision pneumatic planter for cotton developed at TNAU (Plate 2) has a metering mechanism consisting of disc with metering holes on a particular radius and chamber. As the disc rotates, the vacuum applied to the metering holes through the chamber enables them to pick up seeds from the reserve hopper next to the disc.A three Hp vacuum blower with a maximum capacity of 230m^3/h was used to generate the vacuum pressure for the metering system. Runner type furrow openers were

Plate 2 Prototype precision planter for cotton

provided behind which cast iron pressure wheels with appropriate pressure bars were used in the developed planter. The per cent of multiple, one seed and missed hill were recorded as 2.7, 96 and 1.3 respectively.

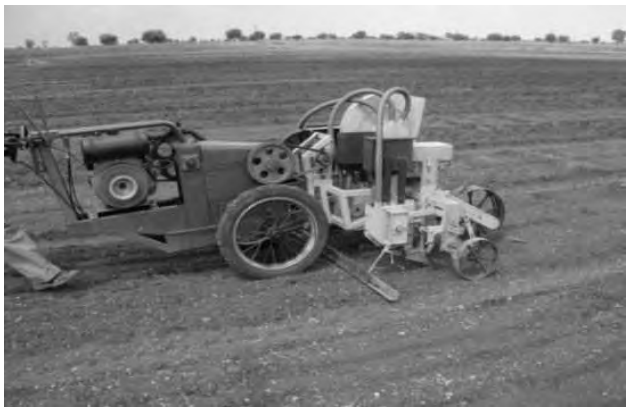

Plate 3 Self propelled pneumatic planter for Cotton sowing

A self propelled pneumatic planter was developed at PDKV as an attachment to the 5 hp petrol engine powered prime mover for precision sowing cotton (Plate 3). A blower with special features of high suction capacity was designed and developed. The blade of the blower was designed to create sufficient negative pressure.The soil moisture content was 27 per cent at the time of sowing and found equipment working satisfactorily in high moisture content.The travelling speed was 3.2 km/h which was suitable for operator to operate for two hours continuously.Actual field capacity 0.51 ha/h with 88 per cent field efficiency.Cost of operation was Rs. 215/ha which is remarkable less than any other traditional method.

Performance evaluation of zero till planter for cotton (Plate 4) was carried out at UAS, Raichur. The zero till planter is operated in the field when the soil moisture is about 24-27 per cent and stubble height of previous harvested crop not more than 15-20 cm generally agreed to be a practice that eliminates seed bed preparation as a separate operation or combines it with planting operation. During the field evaluation, the zero till planter was operated at an optimized forward speed of 3.3 km/h. The soil texture was medium to deep black. The moisture content of the soil during the test was 11.5 per cent. The average depth of seed placement of seed in zero till planter and traditional sowing method is 32 mm and 34 mm respectively. More uniform depth of seed placement was obtained with the zero till planter compared to traditional sowing method. The germination percentage for cotton seeds varied form 65 to 80 per cent. This means that percentage of missing hills using the zero till planter was within the range of germination percentage.The field capacity of the zero till planter was 0.72 ha/hr with a field efficiency of 76 per cent. The fuel consumption was 3.2 lit/h. The cost of operation in terms of Rs/h and Rs/ha is Rs.366 and Rs.508 respectively.

Plate 4. Field evaluation of Zero till planter for cotton in field

A Bullock drawn vertical rotor type cotton planter suitable for planting of delinted cotton seed at specified row and plant spacing in vertisolswas developed at CICR, Nagpur (Plate

5). It was found to have lesser seed requirement, greater accuracy, saving time and labours over the traditional manual method. Two vertical discs with notches on periphery are mounted on shaft driven by a ground wheel through chain and sprocket assembly with transmission ratio of 1:1. The average depth of seed placement was 6 cm below ground with a germination percentage of 84 per cent and a seed rate of 5.2 kg/ha. The field capacity of the implement was 4.5 hr/ha.

Plate 5. Bullock drawn vertical rotor precision planter

In northern India Happy seeder machine is popularly used for direct sowing of wheat in combine harvested paddy fields in standing stubble conditions. An inclined plate planter attachment for planting cotton using happy seeder in wheat residue was developed and tested at PAU, Ludhiana. This mechanism was attached with happy seeder for sowing cotton in standing stubble conditions. The inclined plate planter was designed and developed as a rear mounted attachment to conventionally available seed cum fertilizer drills. This machine can sow bold grains like of soybean, cotton, groundnut etc. Yield recorded for inclined plate planter was 1675 kg/ha and for happy seeder was 1390 kg/ha.

Plate 6 Inclined plate planter attachment to seed cum fertilizer drill

Plate 7. T. O. Inclined plate planter

Weeding and IntercultureEquipment : Weeds reduce cotton crop yields by robbing the plants of moisture and nourishment. There are several ways to control weeds. In many developing countries, farmers remove weeds by hand. Nearly all farmers grow cotton in rows to make weed removal easier.

Traditional mould board plough is found satisfactory in few soils while it creates the problem of dust mulch with severe soil erosion in Sandy loam soils. There is varied response for number of ploughing. However, deep ploughing is recommended once in 2-3 years in Sandy and clay loam soils (Kairon, 1979).

Hoeings are accomplished with bullock drawn weeders and interculture tools like blade harrow, 3 tyne cultivator. Former gave a better performance as compared to latter (Behl, *et al.,* 1989). For later stages of crop growth tractor drawn high clearance cultivators using full and half sweeps give good results. Shovels can be chosen to suit local conditions. Progressive farmers of North zone use Ridger ploughs for row crop cultivations and earthing up operations at advanced crop growth stage.

Plate 9. Animal drawn sweep cultivator

Plate 10. Self propelled inter row cultivator

A self propelled inter row cultivator was developed at PDKV, and tested in cotton and other crops (Plate10). In cotton it was tested in check row of 60 x 60 cm. The speed of operation was 2.85 to 3.2 km/hr. which was moderate for maximum efficiency and drudgery free to the operator. Due to this, operator could work continuously for 3 hours. The actual field capacity of was found to be 0.25 to 0.3 ha/hr with field efficiency varying from 48 to 51 per cent. Similarly, the weeding efficiency was 85 to 86 per cent.

Plate 11. Tractor operated three row rotary weeder

Plate 12. Single row manually operated self propelledweeder

A Tractor operated three row rotary weeder(Plate 11) along with two commercially available self propelled machine used for

weeding purposes *viz.,* RPW 1 with a 4.8 hp diesel engine and RPW 2 with a 6.5 hp petrol engine, riding type (Plate 14) were evaluated at TNAU, for weeding in cotton. Performance of the machines was satisfactory. Weeding efficiency was about 94-95 per cent for both self propelledweeders as compared to 90 per cent for tractor operated rotary weeder. However, the field capacity was less for the former. Injury to plant was 1-3 per cent. Saving in cost and labour was 30-40 per cent and about 90 per cent as compared to manual weeding.

Single row manually operated self propelledweeder(Plate 12) is used for weeding and tillage operation in orchards and in wider row crops. It consisted of a 4.8 hp light- weight diesel engine mounted on the power tiller chassis, power transmission system, two M.S. wheels, a frame and rotary blades. The power tiller and tractor operated weeders, gaining popularity among farmers of row crop were evaluated and modified at PAU for the frequent breakdowns suffered by them. The field capacity of self propelled rotary weeder was 0.07-0.09 ha/h and of tractor operated weeders was 0.25 -0.33 ha/h respectively. Weeding efficiency of all type of row weeders was about 92-96 per cent.

Apart from tine type cultivators rotary blades as well as power operated rotary gauge type cultivators are now available. Significantly tractors with relatively high chassis clearance (upto 75 cm) are being used for operating these cultivators.

Spraying equipment : Pesticides must be used more wisely if we are to reduce the amount of chemical applied and the number of applications, thereby decreasing selection pressure for resistance, prolonging the useful life of each pesticide and reduce environmental contamination and residues in agricultural produce (Southwood, 1977). The role of pesticide application equipment to secure a uniform deposit of the chemical on the target substrate without any wastage (drift or drip) of the material can not be undermined. The main concern being the coverage of the whole area as completely and quickly as possible, little or no attention is paid to droplet size. Smaller droplets are subject to exodrift while the larger ones are the main components of endodrift. A wide range of nozzles are available, most of them producing a wide spectrum of droplet sizes. Since pesticides are most effective in a narrow range of droplet sizes for each specific target, the droplets outside the range only contribute to losses from either endodrift or exodrift. It is recommended that nozzles producing appropriate droplet sizes for particular pests should only be selected.

Table 3. Weeding and hoeing equipments used in India

Sr. No.	Name of Equipment	States where used
1	Mould board plough	North zone
2	Blade harrow	Central zone
3	Cultivator	North zone, South zone
4	Tractor drawn high clearance cultivators	South zone

Most commonly used sprayers on cotton farms are Lever Operated and Power knapsack sprayers. Droplets in the volume medium range of 120-300 micron with 20-50 droplets/cm^2 of leaf were found effective for cotton pest control. Maintaining a constant pressure while spraying improves the production of uniform size droplets. This can be achieved automatically by incorporating a pressure management valve in the lance. It is possible to reduce up to 60 per cent cost of spraying with this equipment if improved methods of spraying are followed. Power

sprayers can be used for quick application of pesticides in dust and liquid form. These sprayers are fitted with two-stroke petrol/ kerosene engine of 1 to 1.5 hp. These knapsack sprayers are very effective at the early stage of crop growth. They pose severe problem at later stage of crop growth since movement of operator as well as handling of lances for spraying becomes difficult. Brushing against the treated foliage contaminates the operator. The performance of power sprayer is better due to higher pressure generated by engine and pump and thereby a strong blast of air strikes at higher velocity and shakes the plants. Power mist blowers spray even the underside of leaves. The field capacity of LOK sprayer varies from 0.4 to 0.5 ha/day while the field capacity of power sprayer varies from 4.0 to 5.0 ha/day.

Vadivelu *et al.,* 1986 studied the efficacy of different types of sprayers (high volume, low volume and ultra low volume) on control of cotton pests and found that with fog air sprayer bad kapas content was lowest, droplet density highest and plant coverage good. They concluded that low volume sprayers are better than high volume sprayers for controlling cotton insect pests.

Plate 13. Self propelled high clearance sprayer

A self propelled high clearance sprayer (Plate 13) for pesticide application in cotton has been developed at PAU.A 20 hp engine operate it having a ground clearance of 120 cm. Swath width is 10.8 m. Machine wheel track is 135 cm, accommodating two rows of cotton crop under the chassis the boom is fitted with 15-18 nozzles and its height can be varied from 31.5 to 168.5 cm. The capacity of machine is 1.6 to 2.0 ha/hr with 200-250 l/ha application rates. Tank capacity is 1000 litres.Labour requirement for the operation of this machine is 1.5 man-hr/ ha.

Plate 14. Air sleeve boom sprayer in operation

Air sleeve boom sprayer (Plate 14), developed and evaluated at PAU is useful for uniform and accurate application and deposition of pesticide and fungicide. It is mounted on 3 point linkage of the tractor and is operated by tractor PTO. The machine is comprised of a 400 liter capacity tank, 18 atomizer, centrifugal blower and hydraulic pump. Atomizers release the pesticide solution into stream of air blast produced by the centrifugal blower. The air blast distributes the chemical in the form of very fine particles throughout its swath width behind the tractor. An isometric view of the machine and view of machine in operation is shown in Plate 18.Field capacity of the air sleeve boom sprayer was 1.70 to 2.0 ha/h and tank filling time was 27 minutes.

Plate 15. Solar powered knapsack sprayer

Solar powered knapsack has been designed and developed at CICR (Plate 15), using a 18 watt solar photovoltaic panel and charge controller mounted on top of a battery operated sprayer. Apart from spraying, the same power can also be used for the household electricity when power supply is not available. The weight of the sprayer without pesticide is 9 kg, with a swath of 90cm giving 10 sprays with a single charge. Manual stroking is avoided which results in reduction of drudgery. At the same time the droplet size generated is uniform due to voltage remaining constant because of simultaneous charging by the solar panel. It overcomes the constraints of unavailability of electricity in the rural area as battery can be charged from solar panel. It has a field capacity of 4 hr/ha.

Picking : The entire cotton crop in India is hand picked by human laborers. Cotton is a labour intensive crop especially during picking and in recent years labor shortages have started occurring during the peak periods. Hand picking operation requires 450-500 man-h/ha. With the result that substantial amount of cost of cultivation is contributed by picking operation. Mechanical cotton picker are also under research and development. Their success will depend on evolving right plant types suited to mechanical harvesting, proper planting geometry, equipping ginneries with pre and post cleaners to handle trash in machine picked cotton, as well as promoting use of cotton pickers on custom hire basis.

Cotton stalk management : The tractor operated paddy straw chopper was evaluated at PAU for cotton stalk management. It consisted of a rotary shaft mounted with blades named as flails for harvesting and chopping the paddy straw. Two counter rows having serrated blades were mounted on the concave of front portion of straw bruising which further assisted in chopping the straw. The stationary view of the machine and an inside view of machine are shown in Plate 25.Approx. 70 to 80 per cent cotton stalks were shredded in size range of up to 20 cm for all three cotton varieties by paddy straw chopper. Field capacity of paddy straw chopper for cotton stalk shredding varied from 0.22 to 0.36 ha/h and fuel consumption varied from 6.6 to 7.66 l/h.

Plate 16. Tractor operated paddy straw chopper

Mixing of standing cotton residue in soil with rotavator was not found very effective however mixing of shredded cotton stalks with rotavator yielded good results as approx. 83 per cent of both the varieties viz. RCH 134 and IT

905 were mixed in the soil.

Plate 17. Flail type chopper cum loader

Flail type chopper cum loader (Plate 17) in a single operation can harvest, chop and load the chopped crop material in the trailer attached to the machine. The blades on the rotary shaft are hinged and are staggered in 4 lines with 7 blades in each line on a horizontal axis perpendicular to the direction of motion. Working width of the machine is 1.80 m. nd view of flail type chopper cum loader for paddy straw cutting and loading is shown in Plate 27.Approx. 70 per cent cotton stalks were shredded in size range of up to 20 cm for all three cotton varieties with Flail type chopper cum loader. Field capacity of flail type chopper cum loader varied from 0.25 to 0.35 ha/h.

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Impact of elevated CO_2 on cotton productivity – A climate resilience viewpoint

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Cotton is an important commercial crop for the economy of India, with recent global position of leading in production ahead of China, and offering livelihood security for the Indian farming community. It is the crop of commerce, history, civilization, symbol of economic prosperity to millions of farmers, being grown in the country on variable land holdings, different planting dates, soil and water holding conditions and largely under rainfed situations. Sustainability of production, overcoming biotic and abiotic stresses, meeting requisite quality standards and maintaining economic cost of cultivation are some of the challenges that need continued attention of the scientists, development officials, field functionaries and cotton growers. Climate change has also become a major national issue as well as global concern. More than 65 per cent of Indian populations rely on agriculture for their livelihood, which is directly dependant on climate. With such an importance in national economy for providing livelihood security to 60 million people including all the stake holders of cotton value chain, the climate change and its likely impact on cotton crop is fast gaining importance.

Changes in the major independent variables *viz.,* CO_2, temperature and water to the extent that they actually occur may alter plant growth rates, biomass reservoirs and plant community composition at local, regional and global scales. The global atmospheric concentration of carbon dioxide (CO_2) has increased from a pre industrial level of about 280 to 379 ppm. in 2005, and a value of 770 ppm. (double the current levels) is expected for 2100 (IPCC, 2007). Such an increase in CO_2 levels affects the biology of living organisms, including insects (Yin *et al.,* 2010).

Reports of Inter-Governmental Panel on Climate Change (IPCC) revealed that the earth temperature has already increased by 0.74°C between 1906 and 2005 due to increase in anthropogenic emissions of greenhouse gases. For Indian region under south Asia, the IPCC has projected 0.5-1.2°C rise in temperature by 2020, 0.88-3.16 °C by 2050 and 1.56-5.44°C by 2080 depending on the pace in future development scenario. Study showed that global temperature increase might exceed to the extent of 1.8-4.0°C by the turn of 21st century resulting in anticipated greater instability in agricultural (food, feed and fibre) production (Aggarwal, 2008). The increase in mean air temperature is influencing reduction of snow cover and discharge of river water. The expected rise in temperature in higher latitudes will be much more than at equatorial regions. Amongst the seasons, the temperature increases are likely to be much higher in winter (*rabi*) season than in rainy (*kharif*) season.

Agriculture contributes about 28 per cent of green house gas emissions, primarily due to methane emission, especially in rice cultivation, enteric fermentation in ruminant animals, and nitrous oxides from application of

manures and fertilizers to the soils. Increasing atmospheric concentration of CO_2 at alarming rates (1.9 ppm/year) in recent years than the natural growth rate causes a concern. Although global atmospheric concentration of methane (CH_4) was at 1774 parts per billion (ppb) in 2005 and remained nearly constant thereafter, yet, increase in nitrous oxide concentration to 319 ppb in 2005 from pre-industrial value of about 270 ppb again is a concern (Sankaranarayanan *et al.,* 2010)

Increased concentrations of CO_2 may influence the development of insect herbivores directly or indirectly through the effects of a CO_2-enriched environment on host plant chemistry. Elevated CO_2 generally increases photosynthesis rates, above ground biomass, yield, and carbon:nitrogen (C:N) ratios and reduces N concentrations, thus impacting the production of plant nutrients . In turn, lower foliar N and protein concentrations, which cause reductions in leaf nutritional quality (Mattson, 1980; Johns and Hugher, 2002), increase consumption rates, mortality, and development times, and thus decrease the fitness of insect herbivores. Cascade effects of elevated CO_2through plants are often considered to be responsible for the main impacts on the performance of herbivorous insects.

Many dry regions may experience a decrease in precipitation, while some others will become wetter. Precipitation is likely to increase in all time slices in all months except during December-February when it is likely to decrease. Reports indicated its decreasing trend in south western and central parts of India, while increasing trend in Punjab, western Rajasthan, Gangetic west Bengal and sub Himalayan west Bengal (Ramakrishna *et al.,* 2006). As a result of these shifts, crop performances can be considerably influenced. Extensive warming (by 4°C) in Indian sub continent could cause significant reduction in crop yields (25-40 %) in the absence of adaptation and carbon fertilization (Rosenziveig and Parry 1994) although appropriate adaptations would reduce the magnitude of losses. Since warming was more or less the same throughout India, still some areas in western coastal districts would lose heavily, whereas other districts in eastern states would even benefit slightly (FAO 2000).

Elevated CO_2 : Of the four major green house gases causing a concern regarding the global climate change, CO_2 is by far the most significant one in respect of cotton production. CO_2 in the atmosphere is observed to have increased by about 80 ppm/m^2 of air, since the beginning of the industrial revolution towards the end of the 18th century. The current value is about 398 ppm and the rate of increase is estimated to be 1.8 ppm/year . This increase in CO_2 has profound implications on global warming and shifts in precipitation at regional and continental scale. In general, increase in atmospheric CO_2 increases the quantum of yield produced photosynthetically, net photosynthesis, biomass production and ultimate output in term of grain, seed, oil and fibre. Besides greater output, higher inputs (light, nutrient and water) use efficiency in cultivated crops are expected to be realized; and the same at a much greater pace in C3 plants (cotton, rice, wheat) over C4 plants (maize, sugarcane).

In a typical cotton field, the plants will extract about 11.34 tonnes of CO_2/ha to make the fibre (lint), oil (seed), protein (feed) and other plant parts. Out of it, nearly one tenth that is taken from the air is used to produce cotton fibres and about 0.5 tones is extracted to produce some 0.19 tonnes of vegetable oil. In the process, around 7.9 tonnes of O2 is released back into

the atmosphere. Thus, more than 450 m. tons of CO_2 were removed by cotton plants during whole growing season while more than 36.3 M tones of CO_2 that removed from the air were used to form the cellulose in the fibre and was bound in fibres for a considerable period of time and most of the CO_2 was deposited back as an amendment to the soil through carbon sequestration (return of carbon). Carbon sequestered in the world cotton fibre supply is equivalent of taking 7.25 million passenger vehicles from the highways (Sankaranarayanan *et al.,* 2010). Effect of elevated CO_2 on cotton growth and development is more apparent through significantly greater leaf area and higher net photosynthetic rates associated with lower dark respiration and light compensation point than plants grown in ambient CO_2 (Zhao *et al.,* 2004). Greater assimilation rate of plants grown in elevated CO_2 enables in incorporating 30 per cent more biomass during the first 36 days of growth Higher assimilation is due to higher chlorophyll-a concentration following CO_2 enrichment (550 ppm) than ambient condition even under different moisture regimes. Study showed average chlorophyll content was higher both in the wet (7.1 %) treatment and dry (8.2 %) treatments (Printer *et al.,* 1994b). The results clearly indicated that elevated CO_2 or its enrichment produced higher chlorophyll content and consequently, higher output in cotton plants.

Cotton plants grown under elevated CO_2 atmosphere fixed 16% more CO_2 than the ambient grown plants (Khader et al. 2004). At the onset of water stress, the photosynthetic activity declined from the initial level of 24µ mol CO_2 to a level of 5 µ mol $CO_2/m^2/s$ within six days (Table1). Even diurnal changes in photosynthesis rate were also observed under free-air carbon dioxide enrichment (FACE). Midday net photosynthesis rates of both leaves and canopies were 19-41 % higher in the CO2-enriched plots than in control plots since midday stomatal conductance values of leaves were 13-44 % greater in control plants than in CO_2-enriched plants (Hileman et al. 1994). There was no effect of CO_2 enrichment on transpiration of crop, grown under well-watered and high-fertility conditions (Dugas et al. 1994) although the CO_2 fluxes were significantly higher in the free-air carbon dioxide enrichment (550 ppm) than at ambient level and also higher with wet than dry irrigation level (Nakayama et al. 1994).

Biochemical constituents in plants viz., leaf carbohydrate content were also increased by FACE and the increments were much more pronounced in the stems and roots. Starch and soluble sugars in leaves in FACE tend to be consistently greater than in control leaves. Thus, the significant effect of CO_2 enrichment on starch-accumulating plants is through increase of nonstructural carbohydrate, especially starch, in non leaf storage pools (Hendrix *et al.,* 1994). Although N and protein

Table 1. Effect of elevated CO_2 and water stress on photosynthetic rate ($mmol/m^2/s$) in cotton

Treatment		Days after imposition of stress (d)						**Mean**
		1	2	3	4	5	6	
Elevated CO_2 (650ppm)	Unstressed	25.1	25.4	24.9	25.6	25.5	24.8	**25.2**
Ambient(330ppm)	Stressed	23.7	24.6	21.5	14.3	7.7	5.2	**16.1**
	Unstressed	21.0	21.4	20.8	21.5	21.8	22.0	**21.4**
	Stressed	20.5	21.6	18.5	16.7	9.8	6.5	**15.5**

Adopted from Khader *et al.*, (2004)

concentrations in leaves, stems and roots were significantly lower in CO_2 enriched plants than in control, yet C: N ratios were higher for the free air CO_2 enrichment plants than the control (Huluka *et al.,* 1994) and there were no significant effects of interaction involving irrigation and CO_2. Reduction in tissue N and protein concentration and increase in C:N ratio following CO_2enrichment has important impact in agriculture and natural systems.

Physiologically, leaf water relations in a cotton plant under CO_2 enriched environment was also improved (Bhattacharya *et al.,* 1994). The atmosphere enriched with 550 ppm during the day light hours under full irrigation produced decreased stomatal conductance leading in increased leaf water potential. Under water stress conditions, FACE decreased the conductance throughout the season although the effect on leaf water potential is not consistent. Thus, FACE increased the season long biomass accumulation by 39 per cent under full irrigation and 34 per cent under deficit irrigation. The FACE treatment improved the water use efficiency to the same amount in well irrigated and water stress plots. These were confirmed in many studies also (Radin, 1992).

Free air CO_2 enrichment was also found to increase root dry weight and densities in cotton (Prior *et al.,* 1994). Vertical root pulling resistance, larger diameter taproots, dry weight and volume were also higher under CO_2 enrichment. The development of more robust taproot systems under CO_2 enriched environments may allow for greater carbohydrate storage to ensure root growth for continued exploration of the soil profile to meet nutrient and water needs during peak demand periods (Prior *et al.,* 1995). Evapotranspiration (ET), a better crop water use parameter for water relation studies, was, however, not significantly influenced by CO_2enrichment. This implies that irrigation water use would not have to be increased to produce cotton in a future high CO_2 world. However, if a concomitant change in climate occurs, such as global warming, ET in cotton may change in response to the changed weather condition (Hunsaker *et al.,* 1994).

Cotton plants, grown in elevated CO_2, had significantly higher seed cotton yield over that in ambient CO_2 as increase in harvestable yield by 43 per cent was observed at 550 ppm of CO_2 throughout the growing seasons (Nagy and Hendrey 1994). Similar results were also reported (yield increase to the tune of 40 and 43 per cent by Mauney *et al.,* (1994). and Khader *et al.,* (2004). Here, the increase in biomass and yield is attributed to increase in leaf area, more profuse flowering and longer period of root retention. Boll growth and developmental parameters under elevated atmospheric CO_2 did not affect any of the fibre parameters (Raja Reddy *et al.,* 1999).

Available data from greenhouse and laboratory studies suggests that leaf photosynthesis, crop growth and water-use efficiency of tropical plants might increase at higher CO_2 concentrations. However, under field conditions, abiotic (light, water or nutrients) or biotic (competition or herbivory) factors might limit these responses. In general, elevated atmospheric CO_2 concentrations seems to increase plant tolerance to stress, that include low water availability, high or low temperature and photo inhibition.

Temperature : Tropical plants may be more narrowly adapted to prevailing temperature regimes than are temperate plants, hence expected changes in temperature might be relatively more important in the tropics. Reduced transpiration due to decreased stomatal

conductance could modify the effects of water stress as a cue for vegetative or reproductive phenology of plants in seasonal tropical areas (Hogan *et al.*, 1991).

Cotton requires warm days and relatively cool nights for optimum growth and development. Temperature significantly affects phenology, leaf expansion, internodes elongation, biomass production and the partitioning of assimilates to different plant parts. In the crop growth front, the seedlings were insensitive to rise in temperature from 20/12 to 40/ 32°C during the first 2 weeks of emergence, and after that, they were temperature sensitive (Reddy *et al.*,1995) since 40 and 50 per cent less biomass at 20/ 10°C and 40/30°C, respectively were observed as compared to optimum temperature of 30/ 20°C (Reddy *et al.,* 1992). Biomass of 13, 15 and 43 per cent were partitioned to 'squares and bolls' at 20/10°C, 25/15°C and 30/20 °C, respectively which reflects to some extent slower development at the temperatures lower than 30/ 20°C. Most of the squares and bolls were aborted above 30/20°C. When the temperature increased from 20/10 to 30/20o C, total plant weight increased by 36 per cent. Yet, boll weight was greatest at 30/20 o C, and least at both higher and lower temperatures. Boll growth was more temperature sensitive than vegetative growth . It was concluded from temperature studies that optimum temperature for maximum growth rate of leaves, main stem and fruiting branches was 30/22°C (Reddy *et al.*, 1995). This was also the optimum temperature for the quantum of squares and bolls retained/plant since the number of fruiting branches did not increase above 30/22°C.

Moreover, the plants grown at high temperature regimes lost their reproductive capacity to a greater extent than their ability to produce biomass. High temperature environments were also associated with cotton sterility and boll retention problems. Cotton plants grown from seedlings at 40°C for 12 hrs/ day shed all their squares. Plants grown from seedlings in the natural environment and exposed to daytime temperatures of 30, 35 or 40°C during the fruiting period accumulated 47, 5.7, and <1 per cent, respectively of their mass as bolls. Three week exposure to 40°C for 2 or 12 hrs/day resulted in 64 or 0 per cent bolls, respectively retained on the plants (Raja Reddy *et al.,*1992). Developmental rates, as depicted by the number of main stem nodes produced, were sensitive to temperature at 40/32°C although the number of fruiting branches did not increase above 30/22°C. All flower buds abscise from the plant grown at 40/32°C (Raja Reddy *et al.*,1995). Soil warming affects the rooting system as the soil temperature also increases.

Studies on the effect of temperature in different genotypes (Reddy *et al.*, 1992) revealed that pima cotton (Extra Long Staple) was found to be more sensitive to higher temperatures than the delta type of cotton plants. They reflected this high temperature sensitivity by producing no fruiting branches at 40/32°C, fewer branches at 35/27°C and more branches at 30/22°C, whereas the G. hirsutum plants produced the same number of fruiting branches in all these temperatures. Pima cotton exhibited greater damage to their reproductive structures at higher temperatures than the hirsutum cotton type. The study suggests that high temperature through climate change may affect the ELS cotton programme.

Higher temperatures (ambient plus 2°C) is also shown to reduce the lint yield with an overall average of 7 per cent (Pettigrew 2007). Changes in temperature, however, had a dramatic effect on boll set and fibre properties (Reddy *et al.,* 1999). Fibres were longer when bolls

grew at less than optimal temperatures (25°C) for boll growth. As temperature increased, fibre length distributions were more uniform while fibre fineness and maturity increased linearly with the increase in temperature up to 26°C, but decreased at 32°C. Short-fibre content declined linearly from 17 to 26°C, but was higher at higher temperature. To the contrary, most fibre quality traits were little affected by varying the temperature regimes (Pettigrew 2007).

If the predicted global warming occurs, temperature extremes are likely to be much higher which will have a deleterious effects on the existing cultivars adapted to a moderate temperature. Hence, development of heat tolerant cotton cultivars with matching yields and desired fibre quality shall be a priority area of research.

CO_2 x Temperature : Predicting plant responses to changing atmospheric CO_2 and to the possible global warming by high temperature and their interaction are more important than the sole effect. Although rates of main stem node formation and the time required in producing the first square and first flower were not little influenced by atmospheric CO_2, yet these were very sensitive to temperature. Similarly, carbon dioxide levels did not alter the time required producing nodes; yet, number of branches produced was sensitive to both temperature and CO_2. The larger the number of bolls set on the lower branches of plants grown at high CO2, the larger is the sink for photosynthesis than plants grown at low CO_2. This may explain the reason for the observed reduction in number of fruit at the upper nodes of high CO_2 grown plants. More bolls and squares were produced and retained on plants grown in high CO_2 environments, except that none were produced in either CO_2 environment at 40/32°C (Reddy *et al.,* 1995).

Although higher CO_2increased final leaf size and rate of leaf expansion, yet, the effect was more pronounced at higher temperatures (Reddy *et al.,* 1994). The increase in whole plant leaf area with doubling of $CO_2$was due to small increases in individual leaf sizes and a large increase in the number of leaves on fruiting and vegetative branches. At 720 ppm of CO_2 enriched atmosphere, the plants had about 40 per cent more squares and bolls across temperatures than the 360 ppm (Reddy *et al.,* 1999). Cotton plants showed large responses to humidity and a very high level of CO_2 (700 ppm). In cotton plants, the enhanced dry matter yield due to doubled CO_2 concentration was 1.6 fold greater at low humidity than at high humidity (Wong 1993).

Khader (2014) observed that boll number in cotton plants grown throughout crop growth at elevated CO_2 level (650±50 ppm) increased significantly with 1 f C rise in temperature above ambient(30 bolls/plant) and decreased to 17 bolls/plant with further increase in temperature of 3 ^{f} C above ambient temperature

Nutrient management : FACE (550 ppm) often decreased tissue nutrient concentration, but increased total nutrient accumulation. Under elevated CO_2, field grown cotton was more nutrient efficient in terms of nutrient retrieval from the soil and nutrient utilization in the plant (Prior *et al.,* 1998). This enables more efficient fertilizer utilization, better economic returns for fertilizer expenditures and reduced environmental impact from agricultural fertilization practices in the future. A significant CO_2 interaction with N was observed for total bolls produced ($p < 0.001$) and retained ($p < 0.05$) (Reddy *et al.,* 2004). The bolls produced and retained/plant were significantly higher for the plants grown at elevated CO_2 and N+ conditions

other than the treatments. Plants grown at ambient CO_2 and N+ condition and elevated CO_2 and N condition performed similarly for total bolls produced and retained. Thus, the study suggests the greater possibility in realization of higher nutrient efficiency under elevated CO_2.

Huluka *et al.,* (1994) found less nutrient concentration with elevated CO_2 levels but at sufficient range for all the tested elements except N, which was below the sufficiency. Reddy *et al.,* (2004) observed a similar result of leaf N concentration decreased with increasing CO_2 under low and high level of N. These low leaf N concentrations did not reduce the effect of elevated CO_2 producing higher lint yields and the response being highest for plants grown at elevated CO_2 and high N conditions. It is inferred that future elevated CO_2 will not have any deleterious effects on fibre quality and yield, if N supply is optimum.

Elevated CO_2 concentration and photosynthesis : Photosynthesis in C3 Plants is a consequence of a multi step and complicated process that involves several biological pathways. The Pathways are photosynthetic electron transport system (PETs), in which the light energy is altered into ATP and NADPH; The Calvin Benson cycle that is also known as a photosynthetic carbon fixation cycle in which CO_2 is fixed into carbohydrates, as well as assimilation, transport, and utilization of photoassimilates as the organic products of photosynthesis [Pego *et al.*, 2000; Eberhard *et al.*, 2008; Foyer *et al.*, 2012]. The two important steps, PETs and the Calvin Benson cycle, are under the control of many genes/gene products encoded from chloroplast as well as nuclear genomes. While the products of genes involved in photosynthesis have obvious functions, they operate together within the framework of an extensively coordinated photosynthetic network of genes, regulatory components, signaling factors, and metabolic processes (Nouri *et al.*, 2015). The expression of genes in both cellular organelles is highly variable and affected by a diverse range of environmental factors [Berry *et al.,* 2013]. Many environmental stresses such as drought, salinity, flooding, light, unfavorable temperatures, and its rapid fluctuations adversely affect the process of photosynthetic carbon metabolism in plants. It may alter the ultrastructure of the organelles, change the concentration of various pigments and metabolites as well as stomatal regulation [Pinheiro and Chaves, 2011; Berry *et al.*, 2013]. Several reports indicate that photosynthesis cascades are highly correlated with the accumulation of some important proteins such as ribulose-1,5 bisphosphate carboxylase/ oxygenase (RuBisCO) and other photosynthesis-related proteins [Maayan *et al.,* 2008]. To get insight into the photosynthetic gene expression and regulation under abiotic stresses, OMICS technologies such as genomics, transcriptomics, proteomics, and metabolomics can provide detailed information which can be later applied to improve plant yield potentials. In response to various abiotic stresses, plants continuously need to adjust their transcriptome profile [Gupta *et al.*, 2013].

Transcriptomic and proteomic approaches have emerged as powerful tools to analyze genome expression at the transcription and translational levels, respectively [Woodson and Chory, 2008]. These high-throughput technologies have been extensively accepted to study the expression of certain genes and proteins in response to different abiotic stresses [Blankenburg *et al.*, 2009]. Proteomics, as one of the cutting edge molecular techniques, efficiently deals with the functional molecular

studies. Recently, improvement of techniques for isolation and purification of cell organelles and compartments gave new insights into organelle proteomics [Nouri and Komatsu, 2013]. Photosynthesis in plants is under the control of a complex network of proteins. Four major multisubunit protein complexes, photosystem (PS) I, PSII, the ATP synthase complex and cytochrome b6/f complex are involved in the process [Hippler *et al.*, 2001]. These proteins are greatly affected under abiotic stress conditions.

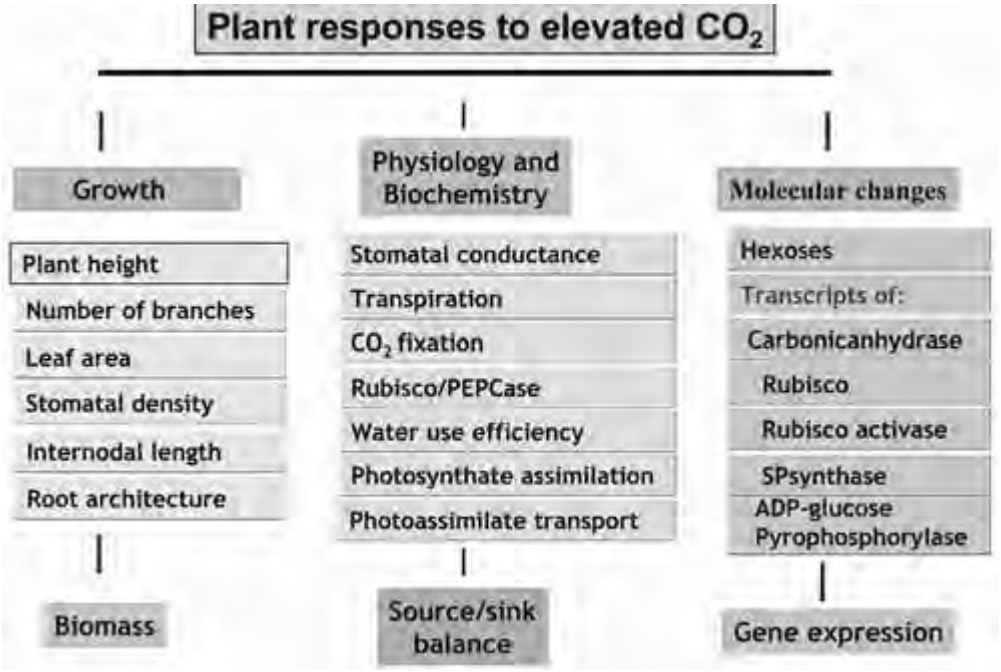
Plants with the metabolic pathways of C3 for carbon fixation are distributed worldwide. They represent over 95 per cent of the earth's plant species, especially in cold and wet climates, usually with low light intensity. In C3 plants, the photosynthetic Carbon Reduction or Calvin Benson cycle for CO_2 fixation produces a three carbon compound, phosphoglycerate. Therefore, plants utilizing this pathway are often named as C3 species [Taiz and Zeiger, 2010]. According to a systems biology analysis, the photosynthetic metabolism of C3 plants has a highly cooperative regulation in changing environments [Ahuja *et al.,* 2010]. Effects of environmental changes and abiotic stresses on photosynthesis system of many C3 plants, from stomatal conductance to carbon assimilation and from gene regulation to protein expression are well documented [Chaves *et al.,* 2009]. Various components are involved in the mechanism of photosynthesis in response to environmental stresses, including photosynthetic pigments and photosystems, the electron transport system, and CO_2 reduction pathways. Changes in CO_2 level of atmosphere is an environmental factor with the most direct and instant effect on photosynthesis. Global atmospheric CO_2 concentration of the earth is 380 iL/L which is 40 per cent more than pre industrial times. Values are predicted to reach between 530 and 970 iL/L by the end of this century [Prins *et al.,* 2011]. In theory, elevated CO_2 will directly affect the balance between photosynthetic carbon fixation and photorespiration. However, plant response to high $CO_2$is under the influence of several factors, including plant carbon fixation pathways. Foyer *et al. (*2012) reviewed the literature related to the C3 and C4 plant responses to elevated CO_2 concentration compared with those grown with ambient CO_2. Exposing C3 leaves to high CO_2, immediately increases net photosynthesis because of decreased photorespiration [Kramer, 1981; Bowes, 1991] and enhances the expression of genes associated with cyclic electron flow pathways. However, long-term elevated CO_2 often decreases photosynthetic capacity, RuBisCO activity and CO_2 fixation.

Crop residue : Assessing the impact of elevated atmospheric CO_2concentration on the global environment is improved by understanding the global Carbon (re)cycling. Carbon fixed within plant biomass ultimately enters the soil *via* plant residues. No significant difference was observed with respect to soil respiration or P mineralization immobilization between CO_2enrichment against ambient CO_2 conditions by application of crop residues. However, significantly greater net N immobilization was observed during the incubation in all soil types with elevated CO_2 treatment by application of crop residue. These results indicate that decomposition of plant residue may not be reduced by CO_2 enrichment, but N dynamics may be markedly changed (Torbert *et al.,* 1995). High CO_2environments without water stress, increased C storage in soil is likely, but it is less likely where water stress is a factor (Wood *et al.,* 1994).

Climate models : Crop simulation

modeling is another approach to predicting the likely impacts of climate change on cotton production in the future (Matthews *et al.,* 2002). COTCO2 model is capable of predicting cotton crop responses to elevated atmospheric CO_2concentrations and potential concomitant changing climate variables. Here, the major plants processes are known to be influenced by CO_2 are simulated explicitly that is photosynthesis, photorespiration, and stomatal conductance, and its role in leaf energy balance. The model explicitly simulates the impact of atmospheric CO_2 concentration on C3 photosynthesis and photorespiration at the level of carboxylation and oxygenation. Growth is simulated for individual organs, such as leaf blade, stem segment, taproot and lateral roots, and fruit, which include squares and bolls. Potential growth is calculated and the carbohydrate and nitrogen required to meet this potential are calculated. Actual growth is based on substrate availability, the potential growth, and water stress. Simulations suggest that if warming is accompanied by higher humidity, the impact of climate change may be minimal. However, if the climate becomes warmer and less humid, ET may increase substantially. Simulations also suggest that enhanced growth due to elevated CO_2 may have a greater impact on ET than climatic change (Zeng and Heilman 1997; Sankaranarayanan *et al.*, 2010).

Tasks ahead : The impact of elevated CO_2 and global climatic changes will have profound effect on agriculture through direct and indirect effects on crops, both through biotic and abiotic routes. Besides ameliorating global climate change through biophysical means (for reduction in emission of greenhouse gases), the issues concerning the global climate change can be tackled to some extent by crop adaptation strategies. CO_2 elevation and associated changes



(Fig. Source: Reproduced from Reddy *et al.*, 2010. CURRENT SCIENCE, VOL. 99 (1), JULY 2010)

are taking place gradually, plants may get adapted to these changes (Hebbar *et al.*, 2007). Rainwater management technologies would play a greater role in rainfed cotton cultivation to mitigate the effect of higher ET by ever increasing temperature and changing pattern of precipitation. By adopting altered date of planting, selection of heat and drought tolerant genotypes, improved land use and natural resource management policies, improved risk management strategies like early warning system and crop insurance and approaches to increase soil carbon, such as, organic manures, minimum tillage, residue management and integrated pest/weed management should be encouraged to reduce the impact of climate change.

Reddy *et al.,* (2010) clearly indicated that increased or decreased biomass yields in plants grown under elevated CO_2 would certainly depend upon the source–sink balance which in turn would be associated with changes in activities of key photosynthetic enzymes and the expression of photosynthetic genes. The morphological, physiological, biochemical and molecular responses of different plants to elevated CO_2 suggests that photosynthetic acclimation and the resulting down-regulation of plant metabolism is due to imbalances between the source sink capacity. Future genetic studies on sugar management for biomass production in green plants, exposed to increased CO_2 concentration in the atmosphere shall be essential (Reddy *et al.*, 2010). Genetic transformation of plants for efficient nitrogen assimilation under elevated CO_2 is also suggested in improving the capacity of nitrogen sink to mitigate excessive accumulated sugars. It would also be useful to understand the impact of elevated CO_2 on primary photosynthetic reactions including photosystem II photochemical performance. Reddy *et al.,* (2010) have also suggested that genetic manipulation of crop plants for positive acclimatory responses is an extremely useful strategy to obtain optimal crop yields under predicted changing global climate.

The interactive relationships of the environmental variables like temperature, radiation, water availability, visible and ultraviolet sunlight, salinity, soil nutrition etc., complicate the predictability of consequences of rising CO_2 in atmosphere (Reddy *et al.,* 2010) . Therefore, the interactive effects of multiple environmental factors on plant responses to rising CO_2 require a careful study. Such

Table 2. Effect of water stress and elevated CO_2 atmosphere on morphological attributes of cotton

Character	Elevated CO_2		Ambient CO_2		CD at (p=0.05)
	Unstressed	Stressed	Unstressed	Stressed	
Plant height (cm)	45.2	38.3	37.6	29.7	2.8
Sympodia number	17.8	17.2	17.4	16.0	NS
Node number	21.4	20.0	20.8	18.2	0.7
Leaf number	64.6	52.0	54.4	48.2	3.2
Boll number	13.8	11.8	10.6	6.6	0.6
Single boll weight (g)	2.08	1.97	1.88	1.76	0.31
Yield (g) /plant	27.4	23.2	20.0	11.6	1.5
Total biomass (g)/plant	83.5	69.0	61.7	39.4	2.8
Harvest index	32.7	33.5	32.3	29.5	1.9

Adopted from Khader *et al.*, (2004)

information should demonstrate how the multiple environmental factors, when altered in a changed climate, could interact with each other resulting in increase or decrease in the growth and metabolism of several plants. Optimization of sustainable and natural fertilizing sources including nitrogen fixing crop rotations, compost, composted manures in cotton production have been suggested for adaptation to climate change (Ton, 2012).

Hake (2012) has suggested further research into adaptation aspects like stress tolerance traits and germplasm, fibre yield enhancement, nitrogen use efficiency, site specific monitoring and input management, phenotypic breeding for elevated CO_2 environments and innovations in fibre quality.

Since climate plays a major and critical role in all the bio activities involving crops, animal and microbes, a concerted effort is needed in forecasting the likely affected parameters of climate change, its probable impacts and its mitigation strategies for fulfilling the cherished goals of sustainable agriculture, particularly a commercial crop like Cotton, and uninterrupted growth/progress of all the stakeholders.

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Recycled cotton fibre as reinforcement for polymer composites

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India's textile industry is a conventional industry dominated by cotton. According to a recent report by the Ministry of Textiles, India, there are 1834 textile mills with an installed capacity of 37 million ring spindles, 489,718 rotors and 56,526 looms. Textiles industry, which includes the nascent technical textiles sector, contributes 4 per cent to the GDP and 14 per cent to the industrial production. The two main reasons which make the Indian textiles industry strong are: export earnings and employment opportunities. India's textiles industry employs some 35 million people directly and contributes 17 per cent to the total export earnings of the country.

Cotton plays a key role in the National economy in terms of generation of direct and indirect employment in the Agricultural and Industrial sectors. Textiles and related exports of which cotton constitutes nearly 65 per cent account for nearly 33 per cent of the total foreign exchange earnings of our country which at present is around 17 billion dollars with a potential for a significant increase in the coming years. Cotton is cultivated in three distinct agro ecological regions (north, central and south) of the country and accounts to 40 per cent of the total global fibre production and is the most important fibre in the world.

Each part of cotton plant is used for various purposes: Cotton linters are used in making banknote and security paper, tea bags, filters, sausages casing and electrolyte condenser. Cotton stalk is used for making particle boards, preparation of pulp, paper, hard board, corrugated boards, boxes and for growing edible oyster mushrooms. Cotton seed is used for animal feed. In India cotton is used extensively for apparel purpose. It has many qualities which make it suitable for apparels such as absorbency, strength, easy spinability, washability, good conductor of heat which leads to comfort in wear during hot weather.

A significant amount of fibrous waste from the textile industry during textile processing and post consumer product is disposed worldwide in landfills each year. That not only poses economic and environmental problems to the society but also represents a severe waste of resources. For economic and environmental reasons, in recent years, increased emphasis has been placed on reusing techniques for various fibrous waste products from textile industries and accordingly research organizations and industries are now looking for applications where waste materials may represent an added value material in composites. Though a variety of technologies have been developed in response to customers demand for recycled products and as alternatives to land filling, the cotton pipeline has a significant environmental responsibility to fulfill.

Today, recycling has become a necessity not only because of the shortage of any item but also to control pollution. There are three ways to reduce pollution. One is to use newer technologies that pollute less. The other is to effectively treat the effluent so that the final

effluent conforms to the expected norms. The third and the most practical way is to recycle the waste several times before it is discharged.

Recycling : Recycling the solid waste may be defined as the recycling of material and its reuse which could include repair, re manufacture and conversion of materials, parts and products.

A huge amount of waste is generated by the textile and clothing industries so there is pressing need to recycle these waste. There are some facts which depict the waste from textile and clothing industry in figures:

- 10 20 per cent of all textiles in the fashion industry are estimated to be wasted.
- About 15 per cent of fabric intended for clothing ends up on the cutting room floor. This waste rate has been tolerated industry wide for decades.
- Over 70 per cent of the world's population use second hand clothes.
- The world supply of used women's clothing is at least seven times that of men's.
- Using recycled cotton saves 20,000 liters of water per kilogram of cotton, a water intensive crop.
- Up to 95 per cent of the textiles that are land filled each year could be recycled.

Textile waste is in pre or post waste stage:

Pre consumer waste is a material that is discarded before it is ready for consumer use. Pre consumer textile waste usually refers to waste by products from fiber, yarn, textile, and apparel manufacturing. The pre consumer textile waste in India has a number of applications based on the fibre composition. Cotton waste is also exported to other foreign countries from India after it is cleaned and the required standard is attained. India is an exporter of refined cotton waste from comber noil and card, yarn waste, hosiery waste to countries like England, France, Malaysia, Thailand, China, Taiwan, Hong Kong and Singapore. Few Indian manufacturers export quality certified recycled dyed and mélange yarns and recycled fabrics from cotton and polyester. Industries like these produce contamination free waste that has varied applications across key industries.

Postconsumer textile waste usually refers to any product that the individual no longer needs and decides to discard due to wear or damage and normally includes used or worn clothing, bed linens, towels, and other consumer textiles. Post consumer textiles wastes are also up cycled in small Indian clusters. Traditionally, fabric from old cotton *sarees* are made into layers and stitched together using run stitches, to give a unique design effect. This product termed as "Kantha" is used for infants and children as blankets and wraps as it is soft and suitable for the Indian climatic conditions. *Kantha* work is famous in the eastern states of India like Bihar, West Bengal, Assam and Orissa.

Recycling of textile waste gives fibre a second life in a rejuvenated life cycle and thus increases the total value of the fibre. Much recycled fibre ends up in low value products. The development of new, higher value products from recycled fibres will encourage utilization of the fibres and contribute to the future sustainability of the cotton industry. Therefore, the reuse and recycling of fibers provide environmental and economic benefits:

- Reducing cost of purchasing materials.
- Increasing profitability.
- Minimizing costs of disposal and

treatments.

- Minimizing environmental impacts by reducing use of new raw materials and producing products from earlier one.
- Textile recycling requires less energy than any other type of recycling.
- Textile recycling does not create any new hazardous waste or harmful by products.

This is in conformity with the "**out of sight out of mind** philosophy. However, recycling is currently accepted as a sustainable approach to solid waste management. Recycling of material from solid wastes help the community economically, environmentally, socially and ecologically. It is a key concept of modern waste management. Recycling is the reprocessing of waste materials into new or reusable products. Ninety nine percent of used textiles are recyclable.

There has been increasing demand for use of recyclable and or biodegradable composites for automotives, especially due to the recent European Union directives. With the growth of automobiles in the global market, and a simultaneous pressure to address the issue of sustainability, there is continual need for the incorporation of natural fiber based materials into automotives

Composites : Fiber reinforced composites (FRCs) have been used for a long time in structural and semi structural applications. FRCs have the advantage of lightweight and best property performance compared to traditional materials such as metals. In several instances, disposability of such products becomes a major issue.

A composite can be defined as a material having two or more chemically distinct phases, which at the microscopic scale are separated by a distinct interface. We can also say that a composite is a combination of properties of two or more components held together by the same type of matrix. In essence, a composite is a commodity having superior properties than the individual constituents. Composites, the wonder materials with light weight, high strength to weight ratio and stiffness properties have come a long way in replacing the conventional materials such as metals and wood.

Composites can be mainly categorized on the basis of matrix and type of reinforcement. The classifications according to matrix type are: ceramic matrix composites (CMC), polymer matrix composites (PMC) and metal matrix composites (MMC). The classifications according to type of reinforcement are particulate composites (composed of particles), fibrous composites (composed of fibres) and laminate composites (composed of laminates). Fibrous composites can be further sub divided on the basis of natural/ biofibre or synthetic fibre. Biofibre encompassing composites are referred to as biocomposites. Biocomposites can be again divided on the basis of matrix *i.e.* non biodegradable matrix and biodegradable matrix.

Natural fiber composites/ bio composite : The bio composites referred to herein are composites that combine natural fibres such as cotton, kenaf, jute, hemp, and sisal with either biodegradable or non biodegradable polymers. In terms of the reinforcement, this could include plant fibres such as cotton, flax, hemp and the like, or fibres from recycled wood or waste paper, or even by products from food crops.

The usage of natural fibres as reinforcement for composites application is receiving increased attention. Natural fibres are lighter, less expensive, have superior specific

strength, require comparatively less energy to produce, good for the environment, biodegradable. Natural fibers combined with traditional plastics offer good performance at lower prices creating huge potential to replace competing materials in automotive applications where even a fractional weight saving can make a significant contribution to energy savings with reduced gasoline consumption and with added advantages of eco friendliness. Natural fiber production has lower environmental impact compared to traditional synthetic fiber production. The light weight natural fibre composites can help in improved fuel efficiency and reduced emission in the use phase of the component especially in automotive applications. As well, end of life incineration of natural fibres results in recovery of energy and carbon credits.

Use of recycled cotton for making composite materials : The composites reinforced by the recycled fibres show similar mechanical properties as those reinforced by virgin plant fibres such as cotton and flax that are traditionally used in composites. Natural fiber composites have gained considerable value in the current market because of their "green" recyclable tag as well as the set of engineered properties they present. Since the early 1900's researchers have attempted to make composites from polymers reinforced with cotton and paper. Only recently the natural fiber reinforced polymer composites have gained more interest and importance than the traditional glass and mineral reinforced composites due to the increasing environmental pollution and stricter environmental policies. Glass fiber or minerals increases the density of the composites, and as such the part specific weight, leading to an increase in energy consumption especially in the automotive and transport industry and not self decomposing or biodegradable which hinders their disposal at the end of their life cycle poses a serious environmental problem. The incineration as well as land dumping of these composites causes air and land pollution which is not desirable in the present or for the future. In view of these concerns considerable research has expanded in the field of making green composites reinforced with fibers obtained from natural resources. For fiber reinforcement, normally scientists prefer thermoplastic polymeric matrices than thermosets due to the low production cycle, lower cost of processing and high reparability of thermoplastics.

Cotton fiber properties are excellent for these types of products and are compatible with some of the major technologies used to produce them. Spinning is one of the vital industries of India and the 4000 ginning factories around the country produce considerable amount of waste during cotton ginning operation. Most of the mills, recover the useful short fibers from the blow room waste by passing them through willow machines, that in turn leaves a non resalable residue called "willow waste". The scope of the waste from cotton industry extends its products to upholstery cloth, curtain cloths, cover cloths, blanket, towels, shirting, quilts, underwear, carpet, industrial roller cloth, electric cabling, hosiery and in the manufacture of asbestos yarn, paper, linoleum, plastic and regenerated fibers. Focusing on willow waste, it is too short a fiber, to be used for any textile application and thus disposed off in the landfills. An investigation report denotes that, the total amount of willow waste generated in India is about 80, 000 to 85, 000 t/annum, and this obviously needs proper treatment apart from disposal as landfill.

There is good opportunity to utilize low value virgin and waste cotton fibre, cotton stem fibres and even gin trash as the fibre

reinforcement in composites for a range of applications.

Several studies have been carried out in order to investigate the mechanical behaviour of textile waste fiber based composites. The usefulness of cotton waste as a source of reinforcing fibers for the preparation of cost effective and biodegradable composites has also been studied. The most common textile waste is denim. Denim is very strong, stiff and hard wearing woven fabric. The fabrics used for today's denim jeans vary. The widely used denim jeans are 100 per cent cotton, 60 per centcotton/40 per cent polyester, 50 per cent cotton/50 per cent polyester and 60 per cent polyester/40 per cent cotton. Some denim used for jeans is a blend of cotton, nylon and polyester. Denim is twill weave fabric that uses colored warp and white weft yarn. We see that denim fabrics have high rigidities, better tear characteristics, moderately sufficient tensile and elevated performance. It is estimated that about 2.5 billion yards of denim is produced every year all around the world. Approximately 1600 pieces of denim will divert 1 ton of waste from the landfill, when recycled. The weight of 1.2 meters of denim is approximately 1.0 kg; therefore the wastage factor for the garment manufacturers is 253 kg for 4000 kg of material processed, or 6.3 per cent. So, the recovery and reusing of denim wastes help to maximize the economies and opportunities by applying this to the supply chain from denim manufacture through to the finished garment. In current researches, waste denim fibers have been used as the reinforcement element to develop a new set of polypropylene matrix composite (Haque, 2014).

Agricultural fibres such as cotton has been used to produce hardboards and have shown better properties compared to the rice straw composition boards. The following are the advantages if cotton stalks are used for the purpose of preparing composite boards:

- Additional income to farmers
- A new material for composite board industry
- Avenues for setting up rural industry
- Employment opportunities for rural youth
- Conservation of forest resources

Cotton has long been a dominant natural fiber in the textile industry. Low quality greige fibers or low value textile wastes predominantly consisting of cotton fibers that could not be used directly in the apparel industry, have a high potential in the manufacturing of composite nonwovens that are quite promising materials in the insulation market, especially in the automobile insulation market.

Use of cotton blends in composite materials: The demand for natural cotton fibres and poly/cotton blend fibres have increased significantly in the past decade Novolac type phenolic composites reinforced with jute/cotton hybrid woven fabrics has fibre orientation and roving fabric characteristics. Jute fibre promotes a higher reinforcing effect and cotton fibre avoids catastrophic failure. Therefore, this combination of natural fibres is suitable to produce composites for lightweight structural applications.

The thermal diffusivity, thermal conductivity and specific heat of Jute/cotton, sisal/cotton and ramie/cotton hybrid fabric reinforced unsaturated polyester composites showed a particular behavior, with thermal properties very close to those of the resin matrix.

Bio composites have been used widely for making building products such as window, door, siding, fencing, roofing, decking, and so on. Bio composites have been classified with respect to their applications in building industry into two

main groups: structural and non structural bio composites. A structural bio composite can be defined as one that is needed to carry a load in use. For instance, in building industry, load bearing walls, stairs, roof systems, and subflooring are examples of structural bio composites. Structural bio composites can range broadly in performance from high performance to low performance materials. A non structural bio composite can be defined as one that is not needed to carry a load in use. Materials such as thermoplastics, wood particles, and textiles are used to make this kind of biocomposites. Non structural bio composites are used for products such as ceiling tiles, furniture, windows, doors, and so on.

India is basically an agricultural country. Around 70 per cent people make their living on agricultural related work. But in past few years there is a rapid migration of population from villages towards big cities. This has caused social imbalance along with crowding of cities thereby putting thrust on basic amenities in big cities, namely food, cloth and shelter. Demand of shelter has given rise to demand of building materials like bricks, cements, steel, etc. Since, the production of building materials is limited, their prices are sky touching and due to fast production, quality is being also suffered .Bricks made up from paper mill sludge and cotton waste can reduce the above problem to some extent by using this composite bricks. This will also produce economical building material since the waste is reused which would otherwise have been wasted. Recent trend indicates that there is a continuous doubling of rates of building materials in a span of five years. The composite bricks which made from paper mill sludge and cotton waste have many advantages like Economical, Environmental friendly, equal compressive strength, water absorption and bulk density as compared with traditional clay bricks.

Applications of composite materials : Newer or alternative composite materials are considered to achieve cost effectiveness, fuel efficiency, reduced emissions, increased safety, and always with a target on future ability to recycle or biodegrade. Lightweight construction, resulting in reductions in fuel consumption and emissions, has been a factor in the move to composites for all transport vehicles. These composites are being used increasingly in:

- Automotive Parts
- Furniture
- Building
- Packaging Materials

CONCLUSION

Textiles in India are recycled both for the domestic and the global market. In the domestic market recycled textile products are generally found in the form of floor mats, wipes and rugs. The fibres extracted during recycling of clothing are converted into recycled yarns and it is used in different textile products and also as fillers. The pre consumer cotton wastes are a source of raw material for the paper industry. Today, in the world of modern technologies, the demand for production is increasing so rapidly in all aspects of the required living commodities. In order to meet all the required demands, over production and utilization of all resources seem not enough. Therefore, the increasing demand for textile making huge clothing production is not only based on demand for more population but it's also changing new fashion habits as well. Improving raw material exploitation has become the most important challenge facing scientific and industrial community. Textile production wastes are undesirable but inevitable by

products in many manufacturing process (spinning, weaving, knitting, or garment manufacturing) and are frequently undervalued. However, if one can convert such wastes into useful product economically, there will be great contribution to the market. Bio renewable resources offer an almost limitless supply of renewable and potentially sustainable raw materials for the production of biocomposites. Although in its infancy, there is a growing market for biocomposite based products and with further development a whole host of new applications can be envisioned. There is a huge range of potential reinforcing fibres/fillers and an extensive range of processing options to ensure the right fibre at the right price.

A large amount of textile waste is disposed of in landfills each year. That not only poses economic and environmental problems to the society but also represents a severe waste of resources. Although the environmental awareness of the general public has increased significantly in recent years, still their willingness to actively participate in waste reduction by recycling needs to be enhanced. So people those are impetuous for waste disposal would think rationally about the rejuvenation of waste fibers to raise the profit

Limitation of recycling :

- Promoting waste avoidance on purely environmental reasons may not be sufficient. As costs for collection and disposal services are not linked directly with the quantity and sorting of wastes generated through charging,
- There is no financial incentive for waste producers to reduce waste.
- Low values, high transportation cost or lack of market demand for recovered materials particularly.
- The predominance of small and medium recovery and recycling enterprises discourages investments in waste recovery technologies.
- The environmental benefits gained from using recycled raw materials rather than virgin materials to make these products include conservation of natural resources as well as reduced energy consumption, carbon dioxide (CO2) and other emissions, and waste going to landfills", .
- There is a good opportunity to utilize waste cotton fibre, cotton stem fibre and gin trash as fibre reinforcement composites for a range of applications.

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Cotton and its prospective uses

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ABSTRACT : Consumers worldwide are becoming aware of natural fibers and slowly but steadily the trend is again shifting to better natural products. Cotton is a natural fiber bestowed with special properties which includes hypo-allergic, soft hand, can be blended with other fiber and many more. Cotton plays an important role in the Indian economy as the country's textile industry is predominantly cotton based. Products developed with the cotton fibers can pave the way to stay ahead in the competition wherein the demand for organic and natural fiber products may even surpass that of synthetic fibers. The innovative ideas and quality functional finishes imparted to cotton fabric and cotton based products would help the growth of cotton in different sectors. Apart from wide use in textiles for clothing, cotton textiles are rapidly capturing the market. Nonwoven, composites, blends made from cotton fabrics are being used for medtech, sportech, hometech, with better performance properties.

Key words : Cotton, Functional properties, Performance uses, Recyclable cotton products

From blue jeans to baby wipes, from dollar bills to dynamite, cotton is more than just the fabric of our lives. Cotton is a natural fiber that comes from the seedpod of the cotton plant. It is a warm climate crop and is mainly grown between 37^0 N and 32^0 S. The northern hemisphere accounts for about 90 percent of global cotton production(International Trade Centre). Cotton has been cultivated and fashioned into fabrics for at least 7,000 years. Archeologist have found cotton fabric fragments in Mexico dating to 3,500 B.C., India to 3,000 B.C., and in the southwestern United States to 500 B.CNot only, all parts of the cotton plant economically useful, but the multitude of uses it can be put to, makes it India's one of the value-added crop. When we think of cotton there is an immediate association with apparel, but there is so much more. An example is paper currency, which is made of 75 percent of cotton and 25 percent of linen. Industrial products containing cotton includes wall covering, book binding, zippers, thread, cotton clothwhile the short fuzzy linters provide cellulose for making plastics, explosives, paper products, paddy for furniture and cotton balls. Linters with longer fibers are used for medical suppliers, twine, candle wicks (Wright, 2003). The cotton seed is crushed to produce oil for salad dressing, and the meal and hulls became animal feed. The stalks and leaves are ploughed to enrich the soil. Cotton is so versatile, just about everything one's wear has the potential of cotton being used, from hats to shoes, jeans to shirt (Cotton Incorporated, 2010).

Cotton and Indian textile industry : The Indian textiles industry is extremely varied, with the hand-spun and hand-woven textiles sectors at one end of the spectrum, while the capital intensive sophisticated mills sector at the other end of the spectrum. The decentralized power looms/ hosiery and knitting sector form the largest component of the textiles sector. The close linkage of the textile industry to agriculture

(for raw materials such as cotton) and the ancient culture and traditions of the country in terms of textiles makes the Indian textiles sector unique in comparison to the industries of other countries. The Indian textile industry has the capacity to produce a wide variety of products suitable to different market segments, both within India and across the world (Anonymous, 2015).

Cotton plays an important role in the Indian economy as the country's textile industry is predominantly cotton based. India is one of the largest producers as well as exporters of cotton yarn. The states of Gujarat, Maharashtra, Andhra Pradesh (AP), Haryana, Punjab, Madhya Pradesh (MP), Rajasthan, Karnataka and Tamil Nadu (TN) are the major cotton producers in India. The Indian textile industry contributes about 11 percent to industrial production, 14 per cent to the manufacturing sector, 4 percent to the GDP and 12 per cent to the country's total export earnings. India is also the second largest producer of cotton worldwide. In India during 2013-2014, cotton yarn production increased by two per cent and cloth production from mill and power loom sector increased by 5 per cent and 6 per cent, respectively. Readymade garments were the largest contributor to total textile and apparel exports from India in FY15. Cotton and man made textiles was also major contributor with shares of 31 per cent and 16 per cent, respectively. China is the biggest importer of raw cotton from India. The other major cotton importing countries from India are Bangladesh, Egypt, Taiwan, Hong Kong.Various reputed foreign retailers and brands such as Carrefour, Gap, H&M, JC Penney, Levi Strauss, Macy's, Marks & Spencer, Metro Group, Nike, Reebok, Tommy Hilfiger and Wal-Mart import Indian textile products.(Indian Brand Equity Foundation, 2015 and Anonymous, 2015)

Consumer and their choices for cotton products With the advent of synthetic fibers during the last few decades, it was felt that the natural fibers would be in oblivion. But the demand for cotton remains high because of its suitability on the basis of price, quality and comfort, across a wide range of textile products. Studies reveal that preference for cotton productsand its role in textile and apparel is high among all demographic segments. The majority of Mexican consumers (75%) prefer clothing made of cotton due to its comfort properties. Japanese consumers relate quality to fiber: 80 per cent prefer that clothing they wear the most be made of cotton and cotton blends. Whereas majority of U.S. consumers prefer casual wear like jeans (95%), socks (94%), business wear like dress shirts (79%) and dress parts (63%), athletic wear (71%) and home textile items like bath towels (90%) and sheets (81%) to be made from cotton and cotton blends. Their positive associate with cotton includes comfort, soft, good quality, durable, natural, fashionable and sustainable. (Cotton Incorporated. 2010; Cotton Incorporated 2013)

Functional finishes on cotton : Reforming to compete : It is well known that cotton possesses the unique combination of properties like comfort, handle, absorbency, wicking, and have traditional uses such as apparels, home textiles and many more. With increasing consumer demand for cotton based products and to compete with synthetic fiber based materials, the textile and apparel industry as well as suppliers are making strong efforts to improve the functional properties of textile materials such as cotton by using nanotechnology, which deals with the science and technology at dimension of roughly 1 to 100 nanometers (Srivasramakrishnan, 2015),

microencapsulation technology; which is a technique by which solid, liquid or gaseous active ingredients are packaged within a second material for the purpose of shielding the active ingredient from the surrounding environment. Thus the active ingredient is designated as the core material whereas the surrounding material forms the shell (Dubey, 2009).Another buzz word in this 21st century is the smart materials such as phase change, shape memory materials and conductive materials, which are used to develop next generation cotton based fabrics that can complement the advantages of cotton and man-made fibers. Some functional properties imparted to cotton textiles includes:

Fire proof (flame resistant) apparel, which is suitable for professional uses and provides effective protection against potential risk associated with high temperature and particularly flashover, flash fire, welding exposures. One such example is UltraSoft® fabric, designed with both comfort and safety in mind. It is created with a unique blend of 88 percent cotton and 12 per cent high tenacity nylon to provide soft feel for greater comfort, enhanced wear life and protective performance. In fact, the fabric is engineered to focus the excellent abrasion resistance of the nylon on the outer surface to prolong garment wear life, while the cotton fibers are focused towards the skin to optimize comfort. (http://www.westex.com/fr-fabric-brands/ultrasoft/)

Temperature management: Outlast® fabric line is other advancement, as this technology is designed to provide comfort by regulating the temperature next to the skin, based on environmental conditions. Temperature regulation is achieved by: phase change technology, which is based on a cyclic process in which latent heat is absorbed, stored and released by the encapsulated particles such as silver based finishes (SilverSmat®), as they change from solid to liquid phases or vice versa. This technology is used to produce hosiery products, footwear by Nike, men's shirt with outlast technology is sold at Jos as well as home textile products such as pillows, mattress-tickling fibers, mattress pads, woven blankets made from cotton fibers. (http://www.outlast.com/en/technology/)

Moisture management: Wicking Windows™ is a unique moisture management application for cotton that eliminates the feeling of wet, saturated fabric against the body. Cotton fabrics generally wick well and typically absorb much more moisture than synthetic fibers. During exercise, many fabrics can become overly saturated with perspiration. As the body moves, most wet fabric tends to cling to the skin, irritate or chafing can occur. The Window Wicking™technique transfer the moisture away from the skin to the outer of the fabric, where it can evaporate, keeping the wearer drier and more comfortable. Another technology which transfers moisture and dries faster is TransDRY®. This technology begins by treating cotton yarn's with a special process to make them water repellent and provide better performance than polyester and nylon. The technology provides effective moisture management performance in a variety of product categories including woven shirting, bottom weight fabrics, denim and socks. (Crumbley, Cotton incorporated)

Water repellent: Many water-repellent treatments inhibit a fabric's ability to breathe and transfer moisture vapor effectively. STORM COTTON™ technology does not affect the natural

ability of cotton fabrics to breathe.. Although it repels liquids, the finish still allows moisture vapor to pass through the fabric where it can dissipate into the environment, naturally keeping the wearer more comfortable. Since the STORM COTTON™ technology minimizes the amount of water the fabric will hold, garments dry much faster than untreated cotton, minimizing the amount of time and energy required for laundering. The STORM DENIM™ is another technology of water repellent finish which is applied in garment form thus allows for greater flexibility to apply various garment finishing techniques to achieve the desired styling and appearance of the finished product (Cotton Incorporated)

Self cleaning: New Teflon™ fabric protector Shield and Clean Portfolio is advanced care for a better planet. In apparel, Teflon™ fabric protector fends off soil, stains and spills on wool, cotton, and blends without impacting the fabric's weight, look, feel, color or breathability. Indoors Teflon™ fabric protector makes it easier to keep upholstery, draperies, bedding and linens looking fresh and clean. Outdoors, Teflon™ fabric protector provides continuous protection for awnings and patio furniture cushions (Chremours).

Antibacterial finish: The demand for antimicrobial fibres, fabrics and apparel has increased sharply since the mid-1990s due to growing awareness among consumers due to the importance of personal hygiene and the health risks posed by certain microorganisms. As a result, there is new generation of antimicrobial products on the market which offer protection against a broad spectrum of bacteria and can withstand up to 100 launderings. A good example of the work being done in this area is DuPont's Biowear® materials for protection against bloodborne pathogens. Sol-gel methods have been successfully employed to impart oil/water repellency and anti-bacterial capability to cotton using fluorocarbon polymer/SiO2 and silver nanoparticle-doped silica hybrid materials, respectively(Tarimala*et al.,* 2006;Yeh*et al.,*2007).

Wrinkle resistant: Nowadays aesthetic aspects have an increasing influence on the overall quality of a garment. Fabric appearance is usually characterized by a number of factors such as strength, pilling, abrasion resistance, shrinkage, drape, color and wrinkles. Cotton with all its positive attributes has one major disadvantage of being wrinkles. Studies reveals that this problem can be solved by introducing shape memory materials in textiles. These are the new class of material which has the potential to remember a pre programmed shape, which can help the fabrics to remove wrinkles without being ironed (Gupta *et al.,* 2014).Vasile *et al.*, 2010 conducted a study to develop fabric with body temperature SMA (Shape Memory Alloys) wires embedded in 100 per cent cotton fabric. Shape memory polymers are also reported to impart wrinkle free, shrink resistance, easy to wash quality and good chemical resistance to cellulosic fabrics such as cotton , ramie, linen.

Cotton as conductors: At the Textiles Nanotechnology Center at Cornell University, Professor Juan Hinestroza and a team of researchers have given cotton threads special properties using nanotechnology. The cotton fibers are covered with microscopic particles enabling them to conduct electricity. The applications of this are endless, but they have already used them to sense the heart rate of a person wearing the fabric(Cattermole, 2010)

Sustainable and recyclable cotton products : Organic cotton is the buzz word in today's sustainable environment. Organic cotton clothing, unheard of a few years ago, is now available in many stores and online businesses. Apparel companies are developing programs that either use 100 per cent organically grown cotton, or blend small percentages of organic cotton with conventional cotton in their products. There are a number of companies such as Esquel and Patagonia driving the expanded use of domestic and international organic cotton, thus responding to market requirements for organic products such as sportswear, personal care items, home furnishing, children products(Shishoo, 2015).Indian brands and companies working in manufacturing and retailing of organic cotton textile and garments includes UV and W the company belongs to the Venus Group from Ludhiana, Punjab and is the first in India to engage in the marketing of certified Organic cotton clothing. Other includes Anokhi, one of the oldest brands in market and an expert in block printing with vegetable colour dyes. Bhu:sattva is as connected to the Earth as its name. With an idea to inculcate ethics and sustainability in the entire supply chain, and, thus, ensuring a Fair Trade Concept(Ayala, 2014). Organically grown, naturally colored cotton is also receiving increasing importance due to their eco-friendly character and range from dark tan, brown, khaki, grey and green(Kranthi, 2014).

Recyclable branded accessories and apparel: Eco Smart by Hanes, one of the oldest branded clothing, is the well known trade mark for fibers made with recycled contents. Eco Smart men's black athletic socks contain at least 55 per cent recycled cotton fibers, and the white socks are produced with 15 per cent yarn content. Puma's Bring Me Back program has a great role in the recycling process. Puma's InCycle is a sustainable collection that includes shoes, apparel, accessories and home insulation materials made either by biodegradable polymers or recycled polyester and organic cotton. Biodegradable lifestyle sneaker developed by Puma is made up of blend of organic cotton and linen and sole comprises biodegradable plastic APINAT Bio, which is biodegradable when disposed of correctly. 100 per cent organic denim is made by Volcom with ozone bleaching and laser finishing to reduce environmental impacts caused by conventional chemicals and auxilarries (Muthu, 2014).

Future of cotton in different sectors : Cotton is playing a very important role in the traditional textile and apparel industry but is not restricted to such uses in recent time. The horizon of cotton products have increased due to the advances made and functional finishes added to cotton based materials to improve as well as enhance its performance properties. Technical textiles are the fastest growing area of textile consumption in the world. As per the market survey it has projected an average growth rate of 4 per cent for technical textiles during the period 1995-2005 (Gopalakrishnan). Technical Textiles is a high technology sunrise sector which is steadily gaining ground in India. Technical textiles are functional fabrics that have applications across various industries including automobiles, civil engineering and construction, agriculture, healthcare, industrial safety, personal protection etc. (Anonymous, 2015)All kinds of fibers such as natural (cotton, silk, coir, jute and wool, kenaf), man-made fibers (viscose, polyester, nylon, acrylic etc.) as well as high performance specialty fibers find their usage in technical textiles with more percentage

of man-made fibers due to their inherent advantage of having higher strength and versatility. The current use of cotton in Indian technical textile industry is 6.8 percent. With new advances and its eco-friendly properties of cotton as compared to synthetics will find its way for different applications.T Rajkumar chairman of Southern India Mills Association (SIMA) said that industrialist involved in technical textiles should look forward to invest in cotton based technical textiles as, India produces nearly 39 million bales of cotton every year, and cotton industry is the backbone of the textile sector. Also, around 70 per cent of technical textiles require man-made fiber and filaments and the cost of these fibers and filaments in India is around 20-30 per cent higher, as compared to China and Indonesia (Anonymous, 2014).Different segments were cotton based products are used and exhibit a potential of increasing market demand are listed as:

Sporttech: With increasing worldwide interest and participation in active sports and outer leisure pursuits have resulted in strong historical growth in the consumption of textile materials in sporting and related goods and equipments**.** Sportswear includes apparels with performance enhancement characteristics (moisture management, comfort, elastomeric, soil guard, anti-microbial) as well as sports goods like inflatable ball and sports accessories;astro-turfs, nets rings etc. **(**Shishoo, 2005).Cotton or poly/cotton fabrics find its place in the production of sleeping bags as inner fabric,organic cotton is used primarily throughout canvas shoes and also children shoes. Pure cotton canvas and polyester cotton blended canvas (polyester/cotton 30/70 or 50/50) are the widely used material for making tents.T-shirts, swimsuits and bikinis made entirely from cotton or cotton blends are becoming popular. (ICRA Management Consulting Services. 2010)

Hometech: The fabrics used in home textiles control interior environment of home. Natural fibers such as cotton, wool, linen, etc., are dominant in the home textiles sector with some of the manmade fibers which includes rayon, nylon, polyester, Teflon etc. The share of cotton and man-made fibers, in the production of home textilesaccount for around 38 per cent and 37 per cent respectively.(Anonymous, 2009). Blends of natural and synthetic fibers have also attracted attention of home textile manufacturers to enhance the fabric performance (Das, 2010).Home textiles includes curtain fabric and draperies which are made with solid color plain weave materials in cotton, flax, wool, silk acrylic etc. Blinds, a type of window textile is usually made up of cotton fabric which gives 100 per cent opacity. Sheets and pillowcases materials normally used are 100 percent carded cotton, ring-spun yarn or open end yarn.. Cotton terry blankets (CBI Market Survey, 2008)bedspreads, mattress covers in traditional jacquard patterns are made up of cotton. Towels made of blends like bamboo and cotton, soy and cotton (Soycot®) exhibit better soft as well as absorbency. Milk fiber obtained from cow's milk by using new bioengineering technology is being blended with cotton to regulate the air quality and exhibits properties like soft, brilliant, antibacterial, absorbent and humectants, thus having great potential for household and spa towels, bathrobes, bed linen and bedspreads (Das, 2010).

Medical applications: Cotton is majorly used in the manufacture of Medtech nonwoven products due to its highly absorbent nature. It is naturally breathing; it has good aesthetic

characteristics, keeps dimensional stability and strength even at high temperatures. The use of nonwoven cotton fabric is for hygiene products, which includes disposable diapers, baby wipes, feminine hygiene products, training pants: wipes such as industrial wipes, surgical wipes: medical includes sponges, dressing , contamination control gowns, incubator mattresses, heat packs. Five percent of the Indian technical textiles market comprises medtech applications. Of this, nearly 35 per cent goes into sanitary napkins, healthcare and hygiene products, another 30% comprises surgical dressings and sutures represent around 20 per cent. Medtech also includes diapers and orthopaedic implants (Anonymous, 2006).The most frequently used biotextile, especially in the process of surgery, sutures help close the wound, a function that will last for a relatively long time until the natural healing process is restored to provide a sufficient level of wound strength. Natural materials, including flax, hair, cotton, silk and catgut, had been used for this purpose in many centuries. Wound dressing is another important application where cotton is being used to treat different types of wounds. In line with the vapour transmission rate of the dressings, non-occlusive dressings, mostly based on woven cotton gauze, allow water vapor and fluid to freely transmit into and out of the wound(Zhong, 2012).

Mobiltech: The application of natural fibers in the automotive industry as interior components has been developed since 1995, primarily in Europe. Natural fibers such as cotton, flax, kenaf, and their blends have been explored to develop products that can go into automobiles and find their application in interior parts such as trunkliners, headliners, carpet backings, dashboards, acoustic insulation and absorbent materials.Shift towards the use of natural fibers is due to the new European directive, by 2011, over 95 per cent automobiles should be recyclable (Ramkumar,S)and to reduce weight of automobiles as a small solution to improve the fuel efficiency and help the global warming issue. It was found that the automotive nonwoven developed by using kenaf fiber blended with cotton fibers, recycled polyester and off-quality polypropylene could meet industry specifications of flammability, noise absorbent, odor, biodegradability as well as strength properties (Zhang, 2004).A trans-atlantic alliance involving University of Tennessee-Knoxville, USDA-SRRC and the University of Bermen, Germany has been working towards cotton composites for automotive applications and found that intimately blended cotton fibers with binder fibers gave better composite and revealed that cotton can form a good substitute for synthetics**.** (Ramkumar, S)

Other sectors includes

Agrotech: These are the Agro-textiles, also known as Agrotex, that are used in agricultural applications related to growing and harvesting of crops and animals**.**Mulch & Seed Innovation in Centre, Alabama, is turning the by-product from cotton ginning into high-quality mulch. While most other mulch is made from virgin trees, this mulch is made using cotton gin "trash." The result is reduced waste going to landfills, and mulch production turning into a green industry itself.(Cattermole, 2010)

Geotech: Natural fibers such as jute, cotton, flax are also finding increasing application, particularly in Asian countries (Reddi, 2003). One such example is the production of reinforced composites by using blend of cotton, flax and recycled

polypropylene.(Foulk *et al.,* 2006**).** In erosion control where the short life span of the geofabric is an advantageous property, natural fiber such as jute and coir are used to control erosion of hill slopes and embankment slopes. Cotton fiber perhaps can be tried in this area provided the property of the fabric is engineered to the specific end use.(Rakshit *et al.*, 1994)

Buildtech: Buildtech segment comprises of textiles or composite materials used in the construction of permanent and temporary buildings as well as structures. Textiles are used in construction for concrete reinforcement, façade foundations systems, interior construction, insulations, proofing materials, air conditioning, noise prevention, protection, visual protection, protection against the sun building safety (Anonymous, 2013).Thin composite boards have been manufactured with the noils obtained from the cotton combing process, blow room waste, and polyester resin at room temperature utilizing a compression method. Such composite boards made by using cotton waste show the potential to replace wood and fiber products as the thin boardsfor furniture and interior(Zhang 2004).

Packtech: It includes several flexible packing material made of textile used for packing various goods for industrial, agricultural, consumer and other goods. It ranges from polymer based bags used for industrial packing to jute based sacks used for packaging food grains and packaging used for tea. Cotton is mostly used for wrapping fabric made out of HDPE/PP and cotton canvas. (textilelearner.blogspot.in/2013/01/packtech-textile-packaging-material.htm)

Protech: Protective Textiles are textile products and related materials used in the manufacture of protective clothing for personnel working in hazardous environments. Protective clothing includes garments for protection from harmful chemical environment, extreme temperature environments, low visibility, ballistics, and protection from other types of severe impact hazards. Woven fabrics using conventional fibers such as cotton, viscose and polyester are treated with Flame retardant finish for making of FR apparel and upholstery. Working suits for professional groups that are occasionally exposed to unforeseen flames and heat are also made from fiber blends such as Basofil/cotton and Basofil/wool (Anonymous, 2006).

CONCLUSION

Cotton is a versatile fiber with multiple uses ranging from food to cloths. The use of cotton in different sectors can be increased by blending it with other fibers, by using new methods and processes to produce a durable and advanced fiber material such as nonwoven. Apart from traditional uses like clothing, cotton fibers, yarns and fabrics developed with functional properties such as water repellent , moisture management , soil and stain removal that is self-cleaning, antibacterial performance can increase their share in Protech, Sporttech, Hometech, Medech, agrotech, buildtech, mobiltech, as there is an increase observation of health and hygiene, use of protective clothing in everyday consumer clothing as well as increase in automobiles manufacturing in India.With organic consciousness and increasing market demand for natural products, there is a need to exploit the full potential of the “white gold” of India, as the name given to cotton.

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Cotton production strategy for the next decade by seed industry perspective

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Indian cotton has witnessed dynamicchangesright from cultivation of predominantly diploid varieties in the beginning to cultivation of allotetraploid *hirsutum* varieties to hybrids and now the transgenic era. Each phase had its own reasons and merits for adoption. Although this kept India as leading cultivator of cotton and maintained its position in top three cotton producing countries in the world, on producibility front, we were well below the average world productivity for the obvious reasons of holding size, dependence of major cultivable area on monsoon and adoption of semi-mechanized cultivation practices.

About 70 per cent of global cotton production comes from four countries which includes India, China, USA and Pakistan. Present global area of cotton cultivation area is 34.1 M. ha and is expected to remain between 32 to 35 M. hain the coming decade and cotton consumption is going to increase with the growing population.With fluctuating acreages in cotton growing countries, focus will be primarily on India for meeting increasing demand of cotton.

Climate change is likely to bring a noticeable increase in surface air temperature in future, becoming more conspicuous after 2040. It is likely to bring significant changes in the hydrological cycles. As a result major river basins are expected to experience water shortages. Studies have indicated that enhanced CO_2 levels up to 650 ppm and increased temperatures up to 40^0C have been found to be optimum for cotton crop growth but the climate change is also expected to bring changes in disease and pest dynamics in the crop. (Cotton and Climate Change-Impacts and Options to Mitigate and Adapt, International Trade Center, 2011). These changes are also going to influence competing weeds and will need proper approaches to control them.

India has seen a dramatic change in the productivity levels after introduction of transgenic technology for insect resistance. Productivity of cotton lint close to 322 kg/ ha (2002-2003) has increased to 537 (2014-2015) kg/ha (Annual Report AICCIP) after adoption of the *Bt* technology and has been evenat that level for some time. In India, cotton procurement prices are based on seed cotton. This is unlike other countries where the prices are based on the lint.Ginning turnout is getting a secondary focus as a trait of improvement. The level of heterosis in cotton isnot comparable with crops such as Corn and hence the expected yield gains are not somehow reflected in the production gains of the country.

Cotton continues to face a stiff challenge from man-made fibers in the growing consumer demand. Manmade fibers will grow at an increasing rate than cotton in the coming decade but Indian market is expected to be more cotton centric.In spite of the growing consumer

demand, cotton will be losing the pace of growth to man-made fibers on account of several factors such as procurement prices, reduction in arable land, stiff challenge from food crops for arable land and challenges of cultivation. There is also a need to bring more discipline in cultivation of the crop to ensure better quality cotton in order to maintain the competitiveness of the crop with the man-made fibers.Breeders also need to focus on the requirements of the cotton based industry mainly the apparel industry to identify their needs and breed cotton to meet their expectations.

Hybrid cotton era has really helped the Indian seed industry to reach its current position in the business. Although the seventies and eighties belonged to the public sector in delivering high yielding cotton hybrids in the market, Indian seed industry has contributed to its success by taking on the onus of seed production and distribution in the market. Success stories of H 4, H 6 and NHH 44 are a testimony of this phenomenon. This has also stimulatedseed industry to set up own breeding programs in the country and the result is that the business has become more competitive. Seed industry also owns the credit of popularizing the cotton seed production technologies in the country. Today nearly 10 per cent (estimated) of the cotton seed production is done using genetic male sterility.It has also an important role in introducing and popularizing transgenic technology for insect resistance in the country.

Indian seed industry produces approximately 27,000 M tons of processed hybrid seed every year to meet the market demand. With changes in the agronomy, the seed rates are expected to grow beyond 2 packets (0.450 kg packing) from current 1.65 packets/ac. This increasing demand of the seed in the coming decade have to be met with ever increasing challenges from climate, increasing cost of inputs and labour, somewhat stationary production areas and price cap on the end product. Technology evolution is reducing the breeding cycles and the increasing challenge to the crop is making the breeding processes very complex.

The main challenges foreseen by the industry in cotton cultivation are-

1. Ensuring cotton acreages for seed production in future
2. Seed production, both quality, quantity and value
3. Emerging pest and disease scenario
4. Dawdling technology evolution process

Way out

1. After evolution of *Bt* cotton in the country and good market sentiments, the area under the crop increased by around 30-35 per cent when compared to the non-GM cotton era in the country. This has resulted in spread and cultivation of the crop under marginal soil conditions and low management. Theseconditions adversely affect the production, productivity and quality of the produce. There is a need to identifyand promote alternative cropsthat require lower inputs compared to cotton and let cotton crop be limited to core cultivationand better management areas.
2. Improved agronomies. Agronomic techniques for higher plant population per acre need to be optimized. This will improve the per sq. unit production thereby increasing production in the country. Standardization of the dosages of plant growth regulators and harvest aid

chemicals for use on cotton needs to be done while promoting the concept of high density planting in cotton.

3. More importance should be given to lint recovery in the breeding programs. We need to shift our approach from per acre seed cotton yield to per acre lint yield. This will have a direct impact on the production and productivity of cotton crop in the country. Higher lint recovery in cotton needs to be incentivized in procurement as a step to improve productivity of the country.
4. Strengthen efforts in molecular breeding for native trait discovery for the existing and emerging pest and disease scenario in the country. This will support the crop by way of stabilizing the production. Molecular breeding techniques can also be used to study diversity within the germplasm and improve germplasm resources in the country for yield and quality enhancement.
5. Improving the seed production efficiency in the crop by promoting use of male sterility system.
6. Focus on plant architecture in breeding to meet the future needs of high density crop cultivation.
7. Streamlining GM regulatory system to encourage competition in established traits (IR and HT) and open door for additional biotic and abiotic events.
8. Relax restrictions on germplasm flow. There is lot more germplasm outside India,therefore India would gain more than it loses in exchange. Current “Indianization” of imported germplasm discourages industry from sending new elite material that will not be considered proprietary.
9. Strong post harvest processing facilities need to be developed in light of the mechanized picking operations expected to be adopted in future.
10. Consortium based approach to address major challenges in the crop with equal public private partnership.
11. Breeding approaches to develop specialty fibers to meet the textile industry requirements.
12. Create an environment that allows seed industry to be more market driven to encourage more investment and competition.

A consorted effort with above considerations will certainly help us making bright future for the crop for overall benefit of the farming community and the country.

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New wilt of cotton

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New Wilt of cotton is a challenging problem for plant scientists from all the disciplines. A peculiar wilt like disease was observed in 1978, for the first time in India simultaneously from Adilabad (A.P.) and Khandwa (M. P.) by Mandloi (1979) on the widely cultivated hybrid JKHy 1. Since then it has been reported to occur in moderate to severe form on the other varieties and hybrids in the other parts of the country also. Today, even after twenty five years of its occurrence our knowledge about the malady is fragmentary. The problem has been in the limelight again with the introduction of *Bt* cotton which seems to be comparatively more susceptible to New Wilt as compared to other varieties and hybrid. An attempt has been made to compile the available information on various aspects and presented in the form of review.

Occurrence : The disease was reported from Maharashtra in 1978 from Deccan Canal Area on Varalaxmi in the incidence of 8 – 10 per cent , later assuming epiphytotic proportions (20 80 per cent) in 1983 on DCH 32 , MECH 1, H 6 and RHH 195 (Padagnur, Zirpe, Horde, Ekbote, 1984) and in 1979 from Marathwada ranging between 1 10 per cent in 1982 and 1984 (Mali *et al.,* 1984). Bhale (1984) reported the disease from Maharashtra and Tamil Nadu in 1981 on DCH 32. It assumed serious proportions in Madhya Pradesh and Maharashtra between 1982 and 1984 on MECH 1 (Mandloi *et al.,* 1984; Raj *et al.,* 1984) and in Tamil Nadu (Srinivasan, 1984). Later reports from Andhra Pradesh between 1980 and 1984 indicated an existence of 5 50 per cent (Reddy *et al.,* 1984).

Extensive surveys carried out in M.P. between 1984 and 1987 indicated its severe outbreaks in 1984 , 1986 and 1987 on JKHy 1, H 4, H 6 and MECH 1 (Mandloi *et al.,* 1984; Anonymous, 1985 a, b, 1986, 1987). Surveys in 1989 and 1990 showed an average incidence between 3.0 and 6.2 per cent (Shastry, 1993).Its first report from Gujarat came in 1979 on DCH 32 (Mehta, 1984). Later moderate to severe incidence was observed in 1984 and sporadic occurrence in 1986 (Mehta, 1987).

The wilt was reported on Somnath in Haryana in 1986 and later on with an incidence of about 10 per cent (Raj *et al.,* 1987). Some cases of sudden wilting were observed in 1988 in Rajasthan (Anonymous, 1989).

With the introduction of *Bt* hybrids in 2002 03, reports of large scale sudden drying have been reported from Madhya Pradesh, Maharashtra and Gujarat. In M.P. the problem was particularly severe on *Bt* hybrids in 2002 03 and 2005 06 seasons. The problem of sudden drying of *Bt* hybrids has been discussed elsewhere in this volume.

Loss :

Seed cotton yield/Fibre quality : Wilt is responsible to cause losses in quantity as well as quality on cotton. Losses in yield to the tune of 37.92 and 48.9 per cent have been reported on varieties Laxmi and Varalaxmi, respectively (Anonymous, 1985a, b). Loss in seed cotton was shown as 15.0 and 14.28 per cent on NHH 44 and NHB 12, respectively (Anonymous, 1990). Raj *et al.,* (1990 a, b) observed that a 15.64 to 67.77 per cent reduction in number of bolls corresponded to the 44.75 to 83.54 per cent reduction in yield on a 3 4 months old crop. Similarly, Mayee *et al.,* (1991) observed 49 65 per cent reduction in seed cotton yield due to wilt at different stages of growth. In germplasm accessions 28.9 to 59.6 per cent and hybrids 33.6 to 60.5 per cent reduction in seed cotton yield has been observed. The wilt affected plants show considerable reduction in seed and lint indices, boll weight and seed oil content (Narayanan *et al.,* 1989). The terminal losses as reduction in number of bolls, seed cotton yield on plant basis have been shown to be 43.96 and 44.99 per cent in wilted and 36.58 and 34.58 per cent in wilt recovered plants. The yield losses on field basis were reported to be 1.3 – 4.37 per cent (Shastry, 1993). Wilt is also responsible for reduced seed germination (Mayee *et al.,* 1991; Shastry, 1993). Wilt is responsible for reduction in mean fibre length, fineness and maturity (Ma ndloi *et al.,* 1987; Deshmukh *et al.,* 1989). It was shown that the worst effect of wilt was on fibre maturity (Raj *et al.,* 1990a, b; Shastry, 1993). The lint from wilt affected plants contains high percentage of newters (Jadhav *et al.,* 1992).

Symptomatology : It has been consistently been observed that this disorder remarkably appears at the boll development stage. According to Basu (1984), the wilting is seen in vegetative, square, flowering and early boll formation stages of the crop, the first symptoms appearing when the crop is five to six weeks old. Mali *et al.,* (1984) observed the symptome to appear when the plants were 55 65 days old. Ekbote (1984) and Shastry (1993) have recorded the influence of date of sowing on wilt and found that irrespective of the stage of the crop, wilting occurred in a short span of period. The crop stages, when wilting occurred, were 14 16 , 12 14 and 10 12 weeks respectively. Balsubramanya *et al.,* (1992) found that wilt occurred at the early boll development stage. At Adilabad and Nagpur, the disease was found to appear at flowering and boll development stages (Srinivasan, 1981, 1984; Raj *et al.,* 1984; Bhale; 1984). Jadhav *et al.,* (1992) observed maximum impact of adverse climate in the form of wilt at linting stage. The higher susceptibility of *Bt* hybrid is being linked to the higher boll retention due to efficient bollworm control.

Development of the wilt was closely watched by several workers from initiation of the disease to the stages documenting the exact syndrome. The major steps have been categorized after critically discussing the available picture. Various stages of symptoms of this wilt clearly show that it is different from earlier known pathogenic wilts caused by *Fusarium* and *Verticillium* (Ebbels, 1975). Characteristic symptoms of new wilt have been described by Mandloi *et al.,* (1985) as:

- Sudden drooping of leaves, giving a morbid look to the plant,
- Bronzed green lean surface, reddened petiole and broadening of leaf angle,
- No necrosis and drying of drooped leaves,
- Disorder found as random scattered plants commonly one plant shows wilt out of two at the same bill,
- The root system remains healthy without any internal discolouration,
- The wilted plants revive spontaneously after a pause of 4 6 weeks.

The wilt symptoms were categorized into two groups *i.e.* the slow wilt recognized at an early stage where the leaves become morbid but do not loose their turgidity immediately, turn yellow,

A comparison of symptoms of earlier known wilts with New Wilt at different stages.

Stage	Characteristic symptoms of			
	Fusarium	*Verticillium*	Lubaga	Wilters
Occurrence	Usually in patches, may be large, symptoms becoming less severe towards periphery, appears early, favoured by sandy soils, root knots and warm weather	As small group plants, appears late in the season, favoured by alkaline soils and cool night	As single or small patches with more severity in the centre. plants	As randomly scattered single
Stem	Vascular tissue stained brown, especially immediately beneath the bark	Vascular tissue stained brown usually in scattered bundles through the wood	Vascular tissue stained brown as in *Fusarium*	Vascular tissue unstained, stem becomes reddish externally.
Leaves	Yellow,often parts of the lwaves wilt, shrivel, become brown and fall down	Yellow at edges but do not fall	Yellowing without intervenial yellowing and necrosis	Become bronzed green, petiole reddish and widen the angle with the stem.
Growth	Ceases, infection in young plants fatal, new growth fron lower nodes	Retarded	Ceases.	Ceases or much retarded for 2-6 weeks, no new growth from lower nodes
Cause	Pathogenic	Pathogenic	Non-pathogenic, but sometimes yield *Fusarium equisetii*	Non pathogenic, partly physiological, partly genetic

Several biotic and abiotic agents have been proposed as probable cause of this malady from to time. These are being discussed herein with possible evidences in their support.

reddening starts from margins extending to petiole and stem thus requiring 12 15 days to express complete symptoms; the quick wilt, on the other hand, is typified by sudden drooping of leaves at peak flowering and bolling stage, rapid drying of leaves wilts out pigmentation taking 7 10 days in wilting (Raj *et al.,* 1987, 1990 a, b , 1991). This has been named as parawilt (Mayee *et al.,* 1989) and Adilabad wilt.

These typical symptoms can be distinguished from the similar type of earlier reported wilts by comparing the critical stages described for them as shown in Table 1 (Ebbels, 1975). The disorder has been shown to occur under stress (water stress as well as water logged) conditions (Shastry, 1993). Forced bursting of immature bolls is also observed due to wilt. The disease is prominently seen at flowering and boll development stages of the crop. Most of the workers are of the opinion that one plant out of the two at a bill gets affected in this wilt; however, in rare cases both the plants at a bill show wilt (Mandloi *et al.,* 1984; Srinivasan, 1984; Raj *et al.,* 1984, 1990 a , b, 1991; Shastry , 1993). This has been explained by Shastry (1993) on the basis of probability estimates. The average wilt incidence varies around 4-6 per cent i..e. any 4 6 plants at 50 clibling points will wilt with probability P= 0.0768 0.1128. Further the probability of any two plants (out of these 4 6) showing wilt at the same dibbling point is still lower as P=0.0016 0.0036. This probability will naturally be higher as the incidence level increase.

Biotic agents : The rhizosphere of the affected plants have often been found to be associated with higher nematode poulations Laxamanan *et al.,* (1989) indicated that the disorder could be due to infection by *Rotylenchulus reniformis* . Kurundkar and his associates carried out systematic studies on this aspect for three years and observed that the nematodes reduced the growth of the plants but did not produce the symptoms of New Wilt (Mayee, 2001).

Mali *et al.,* (1984) suggested the need to study the involvement of flagellates in this disorder. Mayee (1993) confirmed the association of flagellates like organism with the phloem tissues of wilt affected plants. However, it has not been possible to typify the nucleus , kinetoplast and flagellum for clearly establishing the presence of protozoal structures. Indirect evidence of protozoal etiology has also been obtained when temporary remission of wilt symptoms has been possible by spraying metonidazole coupled with carbendazm which are known antiprotozoal drugs.

Repeated attempts to isolate a pathogen from the roots of wilted plants of various *arboreum* and *hirsutum* varieties and hybrids were not successful to establish fungi, bacteria and actinomycetes as a pathogen associated with the wilt (Mandloi *et al.,* 1984, 1985 ; Shastry, 1993). Although 10 per cent of the samples yielded Fusarium, Mali et al. (1984) failed to induce typical symptoms. They have isolated a yellow bacterium on nutrient agar from wilting plants; however, its nature as an etiological agent could not be established.

Involvement of viruses has been ruled out by many investigators since the disease could not be transmitted by grafting in situ

(Anonymous, 1987a; Krishna *et al.,* 1988; Mali *et al.,* 1984; Mayee et.al., 1991; Raj *et al.,* 1991 ; Shastry, 1993) and through insects (Jayaswal, 1984; Mayee *et al.,* 1991).The seeds used from previously wilted plants showed poor and inconsistent recurrence of wilt (Anonymous, 1986). Similarly , the recurrence of wilt in the progeny grown from seeds of wilt affected plants was found erratic (Mayee *et al.,* 1991; Shastry, 1993).

Scanning electron microscopy : Vascular tissues of wilting plants remain healthy and unstained (Mandloi *et al.,* 1985). Roots from affected plants do not show any discolouration or clogging (Balasubramanaya *et al.,* 1992).Mayee *et al.,* 1991) through SEM studies have revealed the presence of oval and irregular bodies in the phloem cells of root sections from wilted plants. They considered them to be accumulation of nutrients under moisture stress. Studies of microtome sections of roots, stem and petioles under light microscope (400 X) reveal that there is no apparent blockage of xylem vessels as is known to occur in other wilts. There is no obvious pathological change in the tissues from the affected plants. However, the xylem vessels in affected tissues seem to be of larger diameter as compared to those in healthy tissues (Shastry *et al.,* 1993). They through the SEM studies provided direct evidence of impaired water uptake in new wilt affected plants both under moisture stress and water logged conditions. They observed that emboli were formed in the xylem vessels of roots, stems and petioles of affected plants indicating that the xylem water comes under tension in response to reduced water uptake, the water column begins to rupture and forms gas filled emboli that render the vessels inactive. This seems to create an additional resistance to water transport to shoot. In revived plants, the vascular emboli are either reduced in size or totally disappear indicating that the recovery probably depends on transportation slow enough not to exceed the impaired ability of the vascular system to supply water to the shoot. Vascular repair presumably occurs because tension on the xylem water relieves, the gases in the vascular water redissolve in the nearby water and the water columns reconnect as the gases become fully dissolved.

Nutitional imbalance: The nutrient status of leaves from wilt affected and healthy plants remains unchanged (Reddy *et al.,* 1984); however, it has been observed that immediately after the wilt the uptake of nutrients is reduced in the leaves (Anonymous, 1987, Krishna *et al.,* 1999; Pundarikakshudu, 1984, 1988). The elemental analysis of wilt affected plants by X Ray fluorescence spectroscopy indicate that some inorganic elements particularly potassium and calcium were significantly less in roots of wilted than the healthy plants (Balsubramanya *et al.,* 1992). Shastry *et al.,* (1993) have shown accumulation of N, P, K, Cu, Fe, Mn, Zn and Bo, while the calcium content was reduced in leaves. Similarly, an increase in total amino acids has been observed in seeds, while there was a decreasing trend in leaves. Methionine content tends to remain low both in seeds and leaves from wilted plants. Reversible leakiness and altered cell membrane permeability was

found in wilt affected tissues of roots and leaves. It was proposed that the leakiness and alteration may be due to the transient reduction in the concentration of calcium which permits revival of some plants later when stress is relieved. New wilt was considered to be initiated by an elastic strain emanating from the presence of impaired water uptake/moisture stress.

Physico chemical characters of the soil: Since the pathogenic nature of the wilt could not be established, several workers analysed the physico chemical characteristics of soil and their involvement in the malady. Analysis of the soil samples from wilt affected plots revealed that there remained no appreciable le difference in the composition with reference to macro and micro elements, while the pH of the soil ranged between 8.3 and 8.4 (Krishna *et al.,* 1988). Manganese injury or phosphate deficiency as the cause of wilt was ruled out by Reddy *et al.,* (1984). Further analysis of soil and plant samples suggested that soil nature or nutrition was not the cause of wilt (Pundarikakshudu, 1984). Mudholkar (1984) suggested that certain specific conditions like high pH and sudden waterlogging of soil after a long dry spell could aggravate the incidence of wilt in cotton. Basu (1984) has observed that heavy soils and low soil temperature favour the disease. Shastry (1993) confirmed that physico chemical characters of soil do not play any role in wilt development. Further, soil and foliar application of major (N, P and K) and minor elements (Cu, Mn, Fe, Zn and Bo) failed to control the disease. Soil application of Ca is also unable to revive the plants.

Ecoclimate: Environment seems to pay an important role in the occurrence of the disease (Mandloi *et al.,* 1984; Raj *et al.,* 1984). Mandloi *et al.,* (1984, 1985) and Krishna *et al.,* (1988) noticed that the occurrence of wilt was more associated with weather shocks like dry spell, high temperature or continuous rains for a fortnight during August/September . They found that the occurrence was specifically associated with dry spells. Raj *et al.,* (1990 b, 1991) have, however, observed that prolonged drought was not a pre requisite for disease onset, it plays an important role in aggravating the incidence. An ecoclimate conducive for the development of moisture stress (low rainfall and longer dry spell, higher maximum and minimum temperatures, low soil moisture and higher cumulative rate of change of maximum temperature) during the first 15 days of September is critical for the development of wilt (Shastry, 1993). Higher potential evapotranspiration values i.e. water limiting conditions suggested that PET values could be of use to forecast the occurrence of wilt. Two linear regression models have been developed to relate the occurrence of wilt with cumulative number of dry days and waterlogging duration in days as $Y1 = 0.9362 + 0.1897X1$ and $Y = 1.4013 + 0.7850X2$ respectively, since the wilt develops under waterlogged conditions. Also, through SEM studies Shastry (1993) concluded that under both the situations (waterlogged and dry spell) there was impairment of water uptake and indicated that the principal morphological changes during New Wilt might be of adaptive value in terms of reducing further moisture stress effects under both the situations. These models can be used

to have an idea of occurrence of wilt under prevailing weather conditions.

Role of ethylene: Possible involvement of excess ethylene production with the wilt has been demonstrated by Sahay *et al.,* (1984) and Sahay (1988a). Sahay *et al.,* (1988) and Shastry (1993) have also postulated that the effect of synthetic pyrethroids on the expression of wilt susceptible genotype is mediated through excess methylene production. Artificial spray of ethylene on healthy plants induces wilt symptoms and the plants recover by the spray of silver nitrate due to neutralization of ethylene. Hence, it was concluded that the New Wilt was the result of high ethylene production in the plant (Sahay *et al.,* 1984). Depletion in methionine control both in seeds and leves of wilt affected plants may be due to its conversion into ethylene as is known to occur in stressed plants where methionine acts as a precursor of ethylene . Krishna *et al.,* (1988) have reported the presence of a number of fungi including *Penicillium* spp. In the rhizosphere of wilted plants which are known to produce ethylene in soils. They have suspected their role in excess ethylene production during the progress of wilt. However, occurrence of *Penicillium* spp. In soil is a common phenomenon and their presence alone cannot be attributed as a cause for the wilt (Balsubramanya *et al.,* 1992).

Synthetic pyrethroids and New Wilt: Application of synthetic pyrethroids induces quick wilt like symptoms in susceptible hybrids (Sahay, 1988b; Choulwar and Kharwade, 1989a; Mayee *et.al.*, 1989; Chauhan *et al.,* 1989; Shastry, 1993). Shaw *et al.,* (1989) have observed a linear relationship between the increase in the population of aphids and whiteflies and the incidence of wilt. With the increase in the population of these insects, there was a corresponding increase in the incidence of New Wilt. It is interesting to note that the increase in the population was induced by the application of synthetic pyrethroids. Sahay (1988) and Shastry (1993) have postulated that the iatrogenic effect of synthetic pyrethroids may be mediated through excess ethylene production. Sahay (1988) considered synthetic pyrethroids to be a cause of wilt, while

Shastry (1993) contradicted it because the malady was observed even in unsprayed crop. Mayee, (1992) also found that even after removal of flowers, synthetic pyrethroids sprays increased parawilt incidence disproving the notion that creation of sink due to higher translocation of photosynthate to bolls as a consequence of effective boll retention is responsible for the wilt. In the absence of insecticide too, the wilt does occur indicating clearly that the pyrethroids cannot solely be implicated in the cause of New Wilt but can be an aggravating factor for the disease.

Inheritance: The incidence of New Wilt is very high particularly in hybrids JKHy 1, DCH 32 and MECH 1. The JKHy 1 and DCH 32 hybrids have one of the parents from acclimatized African *Gossypium hirsutum* race *punctatum* as direct parent or in the lineage of one. Some of the hybrids such as JKHy 1 and DCH 32 have Reba B 50 and BJA 592, both from Africa, involved

directly or indirectly as one of the parental lines. In the absence of pedigree for MECH 1. it was difficult to draw inference but could not be different from what is being contemplated (Bhale, 1984; Srinivasan, 1984). In spite of Reba B 50 being used for more than a decade throughout the country, no report on the occurrence of wilt on Reba B 50 casts doubt on this hypothesis. This ais also supported by the fact that the malady has been seen to occur on many other varieties which do not involve any of the African lines in their pedigree. Not only Reba B 50 group but several other genotypes were also found to be affected. Progenies of crosses where these lines have been used also exhibit wilt symptoms. It was proposed that genetic factor alongwith the environment seemed to play an important role (Mandloi *et al.,* 1984, 1985; Raj *et al.,* 1984). Mandloi and Krishna (1988) have studied the pattern of inheritance of this wilt through a 10 x 10 diallel and 12 x 4 line x tester crosses and have proposed a modified duplicate factor model for its inheritance with following assumptions :

- The character is governed by two pairs of independent genes.
- Original population is double heterozygous for this character.
- The factor follows Mendelian laws of inheritance.
- There are equal chances of survival and random mating of gametes and survival of zygotes.
- Penetrance and expressivity of the character is governed by environment.

According to this proposition, the ratio of healthy and wilted plants in a population can be 16: 0, 15 : 1, 14: 2, 12: 4, 8 : 8 and 0 : 16 but the common ratio will be 15:1 as healthy : wilted plants. This , therefore, explains the general occurrence of wilt in the fields around 6.25 per cent. This proposition has been confirmed by Shastry (1993) by studying genetic parameters of variation and showed the role of additive gene action in expression of this wilt. A strong positive correlation exists between the wilt and number of bolls per plant. Higher number of bolls/plant coupled with environmental stress may predispose the plant to wilt. The wilt has been shown to be a genetically controlled physiological disorder expressed under stress (moisture stress or waterlogging) conditions of the environment.

Mathematical modelling to estimate losses : Recently, attempts have been made to estimate the losses in yield due to this wilt. Krishna and his associates have evolved a mathematical model to estimate yield losses as:

$$Y_e = 1 \frac{b_w N_w}{b_h N_h}$$

Where, Y_e = estimated loss in yielded , W = wilt percentage, N_w N_h and b_w b_h are the average number and weight of bolls on wilted and healthy plants, respectively, these estimates are based on the actual number and weight of bolls/plant in healthy and wilted plants and wilt incidence. An improved model has been developed by Shastry (1993) after considering the yield contribution of revived plants as :

$$\text{Potential yield } (Y_p) = \frac{Y_o}{100\, W_1 + W_1 \times 0.58182} \times 100$$

Where , 0.58182 is a standardized constant, Y_o and W_1 are observed yield and incidence of wilt (per cent) , respectively. These models can be used to assess yield losses due to this disease.

Management : The efforts to devise suitable control measures have yielded very little success. Krishna *et al.,* (1988) showed that 60 70 per cent wilting plants recovered to normal by application of 2 3 solution of N+P of 0..5 per cent/wilting plant if drenched as wilt initiation stage. Wilted plants have also shown to recover by application of Thimet + Carbendazim + Metronidazole (Anonymous, 1990) and Metronidazole (Choulwar and Kharwade, 1989a; Mayee *et al.,* 1989; Anonymous, 1992). However, these results did not prove consistent enough to be applicable for field control.

INVITED PAPERS

Genetic enhancement in cotton through conventional and genetic engineering approaches

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Genetic enhancement is necessary and useful but it is not yet well recognized as being so. This is evident because there is no group of scientists or professionals who call themselves "Genetic Enhancers", or "pre-breeders. Genetic enhancement, up to now, has utilized standard hybridization, segregation, and whole plant selection techniques. Back crossing, population improvement, and pedigree selection among selfed progeny are examples of methods employed. But with the advent of molecular genetics and cell biology, a new kind of biotechnology assisted genetic enhancement or pre-breeding is possible. Therefore, unavailable genes for insect resistance or other characters like abiotic stresses may be transferred from alien species into elite genotypes"

The genetic enhancement or pre-breeding refers to the transfer of gene or gene combinations from wild or cultivated sources into breeding materials. The concept emphasizing the use of plant generic resources refers only to the improvement of germplasm. The improved germplasm lines can readily be used in breeding programmes for cultivars development. Therefore pre- breeding does not differ significantly from general framework of plant breeding and is considered as a prior step of sustainable breeding programme. In pre-breeding a useful character is identified from genetic diversity and putting those genes in to useful form. Then the question arises why we need genetic enhancement or pre breeding?

Need for genetic enhancement or pre breeding To meet the ever increasing market demand, plant breeders have to develop cultivars, by using the diverse germplasm lines or elite breeding material. In the past most of the cotton cultivars have been developed through selection and adaptation rather than through creating variability. This has resulted in narrowing down of genetic base resulting in slow progress in plant breeding and increased risk of genetic vulnerability. The best example in cotton is that several resistant/tolerant varieties of upland cotton in north zone like RS 875, RS 810, Rs 2013, F 1861, H1117, H1226 and hybrids like LHH 144, CSHH 198, CSHH 238 and CSHH 243 were developed in north zone by SAU's and ICAR institutes (Ajmera *et al.,* 2004., Tuteja *et al.,* 2005, 2006 and 2009), however, these cultivars have become susceptible over the years due to new strains of begomovirus causing CLCuV (Monga 2014). In order to break these bottlenecks and to create the genetic variability for different characters genetic enhancement or pre- breeding is required to the value of cotton germplasm lines.

Germplasm and genetic enhancement in cotton : Germplasm plays an important role in improving the cotton varieties for resistance to biotic stress and abiotic stresses. Genetically enhanced germplasm lines are also needed for improving the quality characters especially the strength keeping in view the present

requirement of modern textile industry. It can be used for development of early maturing genotypes having synchronous boll bursting, which can fit well for High Density Population System (HDPS) suitable for machine picking. Wild germplasm lines can be used for development of male sterile lines and restorer lines for heterosis breeding programme as well as in broadening the genetic base of cultivars, creating vast genetic variability and in value addition of different genotypes.

(i) Use of exotic germplasm : It refers to all the germplasm that do not have immediate usefulness without selection or adaptation in a given environment. (Haullauer and Miranda, 1981). Exotic germplasm has to go under pre-breeding to find its usefulness in breeding programme. Because most of the plant breeders fear using exotic material due to its detrimental effects on elite breeding material (Kannenberg and Falk, 1995). The major constraints in use of exotic germplasm lines are linkage of undesirable genes with desirable traits is a major constraint to increase the utilization of exotic germplasm, introduction of inferior alleles and disruption of co adapted alleles in elite breeding material. Crossing with exotic material can negatively affect adaptedness when introduced into locally adapted gene base. Therefore exotic germplasm has to undergo conversion or pre breeding to find its best use in breeding program.

(ii) Use of wild species of *Gossypium* genome : The genus *Gossypium* contains about 50 species, including diploids (2n=2X=26; genomes A-G and K) and tetraploids (2n=4x=52; genomes AD1-AD5). The wide geographical distribution of the diploid cottons under primitive or traditional cultivation has provided opportunity for the development of extensive diversity in biotic resistance (Stewart and Robbins, 1994). *Gossypium* species and their interspecific hybrids provide an array of plants that display novel chemistries and resistance characteristics that are relevant to protecting cotton from pests (Bell *et al.*,1994). However, the tetraploid cottons have been the major source of new genes that breeders use, but future improvements in agronomic fitness, quality of cotton and environmental resistance depend on diversity within the genetic resources from which new traits can be selected (Stewart, 1995).

India is the only country in the world where all the four cultivated 'A' genome species known as Asiatic cottons (*G. arboreum* and *G. herbaceum*) and tetraploid species of AD genome referred as New World cottons (*G. hirsutum* and *G. barbadense)* are grown. The *G. hirsutum and G. barbadense* originated in the new world from interspecific hybridization between species of closely related to *G. herbaceum* or *G. arboreum* and American diploid *G raimondii* or *G. gossypoides* (Beasley, 1940). Unfortunately, most of these plants cannot be crossed directly to cotton to make fertile hybrids, but must be genetically enhanced before breeders can use them directly.

Gene pools of *Gossypium* : Three types of germplasm namely, primary, secondary and tertiary based on the fact that the ease with which genes can be transferred from the donor source to the recent parent/species (Harlan and Dewet, 1971). The germplasm pools that are centered on cultivated tetraploid and diploid cotton and various *Gossypium* genetic resources available for improvement are assigned according to biological affinity is given in Table 1. Stewart (1995) assigned the *Gossypium* genome groups to primary, secondary and

tertiary pools based on the ability to generate fertile hybrids between the donor and recipient species, the frequency of genetic recombination between donor and recipient chromosomes, and the ability to produce stable synthesized allopolyploids its segregation gamete formation and viable progenies, respectively.

In the past primary gene pool has been extensively used for genetic improvement of different crops with a view to create vast genetic variability for various traits in cotton. By crossing the *G. hirsutum*, *G. barbadense* and three wild species *G. tomentosum, G. musterlinum* and *G. darwinii* vast genetic variability has been created for morphological and disease resistance traits such as blight resistance, boll worm resistance, *Fusarium* and wilt resistance , cleistogamy and nectriless leaves (Endrizzi *et al.,* 1985 Meredith 1991, Stewart, 1995).

Recently, the work on use of secondary and tertiary gene pool in cotton has been intensified and as a result vast genetic variability has been created for various economic characters. Because the secondary gene pool species are diploids, the initial interspecific F_1 from a direct hybridization with a tetraploid cotton is sterile triploid with a few exceptions (Meyer, 1974) Successful cases for introgression are fibre strength, disease resistance and cytoplasmic male sterility (CMS) and restorer lines (Stewart, 1995). Similarly, the tertiary gene pool represents the most difficult group of species which include E, C, G and K genomes. The only successful gene transfer from this genepool is the introgression of dominant gene controlling terpenoid aldehyde methylation from *G sturtianum* to *G hirsutum* (Bell *et al.*,1994) which imparts natural resistance against insects and microbial attack.

Methods of genetic enhancement or pre breeding : There are two major approaches for genetic enhancement or pre-breeding either through conventional or genetic engineering methods

(a) Introgression: It is a transfer of one or more genes from exotic/un adapted/wild stocks to the adapted breeding population. This is achieved by crossing donor and recurrent parent. This concept of transfer of character through back cross in cotton was first given by Knight (1945).

In back cross method**,** the parent from which desirable genes are to be incorporated is used as donor parent and the parent which is to be further improved is used as the recurrent parent. Six generations of conventional recurrent backcrossing are levels required to transform a genetic stock. This method leads to accumulation of genes resulting in enhanced level of genetic expression of trait. Various type of back crossing methods are recurrent backcross, inbred backcross, congruity backcross and Marker Assisted Backcross Method (MAS) for genetic enhancement of a germplasm lines.

(b) Incorporation: It refers to a large scale programme aiming to develop locally adapted population using exotic germplasm. In contrast to introgression, incorporation aims at indexing the crop genetic base.

(c) Other breeding approaches: There are other conventional approaches like convergent improvement, modified convergent improvement , strain crossing, multiple strain crossing, development of synthetics/composites, decentralized breeding and participatory

breeding for genetic enhancement of a germplasm line

Achievements of genetic enhancement or pre breeding in cotton: Pre-breeding programmes were carried out in USA, China, India and USSR etc during seventies. The important part played by hybridization and introgression in the evolution of new world cotton has been brought out by Hutchinson (1959). The various aspects of gene transfer through introgression in *Gossypium* like disease and pest resistance (Mehetre *et al.,*2002) and fibre quality parameters (Mehetre *et al.,*2003), have been reviewed critically. The potentialities of wild species are given in Table 1.The role of wild species of *Gossypium* as sources of new characters for genetic enhancement has been carried out by several workers and described below:

Improvement in yield: In cotton improvement in yield has been achieved by developing high yielding varieties and interspecific hybrids. In *G. hirsutum* cotton, varieties Arogya, PKV 081, Rajat, Gujarat 67, MCU 2, MCU 5, Deviraj, Devitej, Khandwa 1, Khandwa 2 and Badnawar are derivatives of interspecific hybridization. Commercially cultivated hybrids have been developed both at tetraploid and diploid levels. Varieties like PKV 081 and Rajat have been developed from a cross between *G. hirsutum* x *G. anomalum* (Narayanan *et al.,* 2004.)

Fibre quality traits: The Extra long Staple (ELS) cottons (*G. barbadense*) are known for their superior quality fibres. Besides the ELS cottons, some of wild germplasm also acted as potential source for improving the fibre properties. In Texas, hybridization work involving *G. thurberi* gave successful results in the transfer of high lint strength to upland cotton (Guany, 1952). Kalyanaraman and Santhanam (1955) reported the potentialities of utilizing *G. anomalum* in the transference of low fibre weight with fibre maturity to cultivated *arboreum* varieties. Attempts were also made to utilize *G. anomalum* in the hybridization programme for transference of lint fineness to the cultivated *arboreum* by the above workers and succeeded in isolating some useful types. Marappan (1960) also reported the transference of fineness from *G. anomalum* to the background of *G. arboreum*. A fairly large number of BC_1 F_1 plants indicated the wider scope for recombination and selection of fine linted plants. Similarly Muramata (1969) synthesized hexaploid cotton by crossing *G. hirsutum* and *G. sturtianum* and showed the possibilities of producing spinnable yarn with very high yarn length. Arutyunova and Volkova (1971) attempted the tri-species hybrids by crossing G. *hirsutum* x *G. herbaceum* x *G. harknessi* and recorded very high ginning segregants with 42-43 per cent. Tuteja *et al.,* (2006a) used Introgressed lines as sources for improvement of upland cotton (*Gossypium hirsutum* L.) genotypes for yield and fibre quality traits. Four introgressed lines viz TCH 1648, TCH 1652, TCH 1653 and IH 35 were selected for making crosses with 12 genotypes of upland cotton to explore the possibilities of improving cultivars for seed cotton yield and fibre traits. The cross combinations namely CSH 146 x TCH 1648, CSH 146 x TCH 1652, followed by F 505' x 'TCH 1653', RS 2013 x TCH 1648, RS 2013' x 'TCH 1652' LRA 5166 x TCH 1653, LRA 5166 x IH 35 and F 505 x TCH 1653 showed significantly better performance for seed cotton yield, number of bolls/plant, 2.5 per cent span length and bundle strength.

Table 1. List of *Gossypium* species groped according to germplasm pool by considering tetraploid cotton as cultivated species and having other desirable characters for genetic enhancement

Genepool	Species	Genome	Insect pest resistance	Notes
Primary	*G.hirsutum*	AD_1	*Fusarium* wilt,	Current and obsolete cultivar, breeding stocks, landraces, feral and wild accession
	G.barbadense	AD_2	Blackarm resistance	*Ibid*
	G. tomentosum	AD_3	Drought resistance, thrips	Hawaiian Islands
	G. G.musterlinum	AD_4		NE Brazil
	G. darwinii	AD_5	Bollworms, nematodes,	Galapagos Islands
Secondary	*G. herbaceum*	A_1	Fusarium wilt,	Cultivars, landraces of Africa and Asia Minor, one wild from Southern Africa
	G. arboreum	A_2	*Fusarium* wilt,	Cultivars, landraces from Asia Minor to SE Asia and China; some African
	G. anomalum	B1	Jassid, bollworms, nematodes,	Two subspecies, Sahel and SW Africa
	G. triphyllum	B2	bacterial blight, rust, staple	SW Africa
	G. capitivistridis	B3	length, fibre strength, male	Cape Verde Islands
	G. trifurcatum	B_4	sterility through cytoplasm,	NE Somalia
	G. longicalyx	F_1	Fibre strength, drought resistance,	Trailing shrub, Sudan, Uganda, Tanzania
	G. thruberi	D_1	Bollworms, rust, fibre strength	Sonora Desert, North America
	G. armourianum	$D_{2\text{-}1}$	Jassid, aphids, Heliothis, bacterial blight, gummosis, rust	Baja California (san Marcos Island)
	G. harknessii	$D_{2\text{-}2}$	*Vertcillium* wilt, Fusarium wilt, rust, male sterility through cytoplasm, drought resistance,	Central Baja California
	G. davidsonii	$D_{3\text{-}d}$	Bollworms, bacterial blight,	Southern Baja California
	G. klotzschianum	$D_{3\text{-}k}$		Galapagos Island
	G. aridum	D_4	Male sterility through cytoplasm, drought resistance,	Arborescent, Pacific slopes of Mexico
	G. raimondii	D_5	Thrips, whitefly, bollworms	Pacific slope valleys of Peru
	G. gossypiodies	D_6	Sucking pests	Central Oaxaca, Mexico
	G. labatum	D_7	*Helicoverpa*	Arbores cent, Central Michoacán, Mexico
	G. trilobum	D_8		West central Mexico
	G. laxum	D_9		Arbores cent, Canon del Zopilote, SW Mexico
	G. tumeri	D_{10}		NW Mexico, coastal
	G. schwendimannii	D_{11}		Arborescent, EI Infiernillo Valley, SW Mexico
Tertiary	*G. sturtianum*	C_1	Rust, staple length,	Ornamental, Transcentral Australia arid zone
	G. robinsonii	C_2		Western Australia

	G. bickii	G_1	Bollworms	Central Australia arid zone
	G. austral	G	Lint yield, staple length,	Trans-Australia, north arid zone
	G. nelsonii	G		Central Australia
	G. constulatum	K		North Kimberley (wet-dry tropical), Western Australia Northern NT, Australia
	G. cunnignhamii	K		North Kimberley, W A
	G. enthyle	K		Prostrate, North Kimberley, W A
	G. exgiuum	K		North Kimberley, W A
	G. nobile	K		Trailing, North Kimberley, W A
	G. pilosum	K		North Kimberley, W A
	G.populifolium	K		North Kimberley, W A
	G. pulchellum	K		Prostrate, North Kimberley, W A
	G. rotundifolium	K		North Kimberley, W A
	G. sp. Nov.	K		Arabian Peninsula & Horn of Africa
	G. stocksii	E_1	Staple length, fibre strength, fibre strength, drought resistance	Horn of Africa to Chad
	G. somalense	E_2	Mites, Bollworms	Yemen
	G. areysianum	E_3		Yemen
	G. incanum	E_4		Ethiopia, Somalia, Kenya
	G. benadirense	E		Somalia
	G. bricchettii	E		Somalia
	G. vollesnii	E		
Primitive races				
	Punctatum		Hardiness, resistance to drought, less gossypol jassid, bacterial blight, *Vertcillium* wilt, high lint, strength, fineness,	
	Palmeri, Brasiliense			Sucking pests resistance
	Marie galante		Stem weevil resistance	
	Taxonomic races of *G. hirsutum*			Pink boll worm resistance and high oil content
	Wild accessions of *G. hirsutum*		Boll weevil, *Cercospora* and *Verticillium* wilt resistance	
	Sinense		Wilt tolerance, high fibre length and fineness	
	Cernuuman		High GOT, big boll, long staple, wilt resistance	
	Burmanicum		High fibre length and fineness	
	Bengalense		High ginning, bacterial blight resistance	
	Rozi		Root rot and bollworms tolerance	
	Nadam		Resistance to drought and weevil	

Adopted from Stewart 1995 and Narayanan et al. 2004

Male sterility: The application of CGMS/ Rf system has proved to be an effective means to produce commercial F_1 hybrid seed for many crops (William, 1992). Number of various species imparting male sterility has been reported by several workers. Out of four types of cytoplasmic sources *i.e. G. arboreum, G. anomalum, G. harknessii* and *G. trilobum*, only *G.harknessii* based cytoplasmic male sterility has been more widely accepted and utilized in cotton hybrid program across the world. The first F_1 line of commercial cotton was introduced by crossing an upland cotton (*G. hirsutum*) as a male parent to a wild species *G. harknessii*, (Meyer, 1973). *Gossypium harknessii* Brandagee (D2-2) which is a diploid (2n = 26) was used as female by Meyer (1971) to transfer *G. hirsutum* genome in the cytoplasm of *G. harknessii*. The resultant triploid was made hexaploid (2n=78) using colchicine. Male sterile tetraploid plants were recovered from cross between hexaploid and tetraploids. Another instance of cytoplasmic male sterility was recorded in the diploid Asiatic species (Tayyab, 1982) using the wild species *G. anomalum* as a source for male sterility. It has been reported by Narayanan *et al.,* (2004) that *G. harknessii*, *G. anomalum* and *G. aridum* are the important sources of sterile cytoplasm.

Davis (1979) who studied the A x R and R x B combinations to clearly determine the effect of *G. harknessii* cytoplasm upon the performance of hybrid found no obvious difference between the performance of hybrids carrying either *G. hirsutum* or *G. harknessii* cytoplasm. However, Tuteja *et al.,* (2004) used G. *harknessii* based CMS system in breeding programme and reported that *G. harknessii* cytoplasm source suppresses the yield, ginning, fibre fineness etc. Therefore, the scope of CGMS system will be greater if divergent and stable restorer lines are developed through genetic enhancement or pre breeding.

Development of restorer lines through genetic enhancement: At Central Institute for Cotton Research, Regional Station, Sirsa, 4 CGMS lines viz., CMS SPC 1, CMS SPC 5, CMS SPC 9, CMS SPC 11, were developed using IH 76 carrying *G. harknessii* cytoplasm by back cross breeding. These CMS alloplasmic lines were crossed with restorer euplasmic lines (Cotton Institute Restorers) CIR 7, CIR 9, CIR 20, CIR 26 and CIR 69. The resulting F_1 were selfed to produce F_2. From F_2 onwards the material was handled using Pedigree Breeding approach. The outstanding fertile plants from the segregating generations were selected and selfed and was followed up to F_7 generations. In this way 25 fertility restorer lines namely CIR 97 $P_{1-1,}$ CIR 97 $P_{1-3,}$ CIR 97 $P_{1-4,}$ CIR 97 $P_{2,}$ CIR 97 P_{1-4}, CIR 97 $P_{2,}$ CIR 97 $P_{3-1,}$ CIR 97 $P_{3-2,}$ CIR 97 $P_{3-3,}$ CIR 97 $P_{3-4,}$ CIR 97 $P_{3-5,}$ CIR 119 $P_{1-1,}$ CIR 119 $P_{1-2,}$ CIR 119 $P_{1-3,}$ CIR119 $P_{2-1,}$ CIR 126 $P_{1-2,}$ CIR 126 $P_{2-2,}$ CIR 126 $P_{3,}$ CIR 526 $P_{1,}$ CIR 526 $P_{2,}$ CIR 526 $P_{3,}$ CIR 920 $P_{1-2,}$ CIR 920 $P_{1-3,}$ CIR 926 P_{2-1} and CIR 926 P_{2-3}, were developed which have alien cytoplasm of *G. harknessii* and have the ability of fertility restoration. Parents of restorer lines are indicated in Table 2.These fertility restorer lines were evaluated and characterized on the basis of pollen dehiscence plants were classified as male fertile or male sterile (Tuteja *et al.,* 2006b).

Pest resistance: Cotton crop is attacked by more than 230 species of insects all over the world. In 9 major cotton growing countries 10-15 insects are considered important and 6 species cause major yield losses (Ridgway *et al.,*1994). Therefore the primary objective of genetic enhancement is improving of germplasm lines for resistance against insects by utilizing exotic germplasm. The work of

transferring bollworm resistance from *G. thurberi* and *G. armourianum* to Sakel cotton was reported by Knight *et al.,*(1953). Besides that he also attempted to transfer genes for bacterial blight resistance from a *G. barbadense* to upland cotton, Jassid resistance from *G. tomentosum* and boll weevil resistance from *G. armourianum* were transferred to *G. hirsutum.* Sherif and Islam (1970) derived a hexaploid (F_1 *G. hirsutum* x *G.anomalum* doubled) and backcrossed that to *G. hirsutum*. Backcross derivatives were utilized as promising breeding material for jassid resistance. Introgressive breeding proved its potential for developing disease resistant types as well. Black arm resistance has been transferred from *G. arboreum* to *G. barbadense* and rust resistance from *G. raimondii* to *G. hirsutum*. In USSR, an upland variety namely C 4537 resistant to *Verticillium* wilt was isolated from a trispecific cross.

In India Thombre and Mehetre (1981) reported successful transfer of bollworm resistance from *G. thurberi* to *G. hirsutum.* Vroh Bi *et al.,* (1998) introgressed the 'glandless seed and glanded plant' trait from *G. sturtianum* into *G. hirsutum* using *G. raimondii* as bridging

Table 2. Characteristics of newly developed restorer lines.

S.No	Restorer lines	Characters					
		Leaf shape	Leaf surface	Flower colour	Anther colour	Stem colour	Seed fuzz color
1	CIR 97 P_{1-1}	N	H	CF	CC	GP	White
2	CIR 97 P_{1-3}	N	H	CF	CC	GP	White
3	CIR 97 P_{1-2}	N	H	CF	CC	GP	White
4	CIR 97 P_{1-4}	N	GL	CF	CC	RP	Grey
5	CIR 97 P_2	OL	GL	CF	CC	GP	Grey
6	CIR 97 P_{3-1}	N	H	YR	CC	RP	Grey
7	CIR 97 P_{3-2}	N	H	YF	YC	GP	Grey
8	CIR 97 P_{3-3}	N	H	YR	CC	RP	Grey
9	CIR 97 P_{3-4}	N	H	CF	CC	GP	Grey
10	CIR 97 P_{3-5}	N	H	CF	CC	GP	Grey
11	CIR 97 P_{4-4}	N	H	CF	CC	GP	White
12	CIR 119 P_{1-1}	N	H	YF	YC	GP	White
13	CIR 119 P_{1-2}	N	H	YF	YC	GP	Grey
14	CIR 119 P_{1-3}	N	H	CF	YC	GP	Grey
15	CIR119 P_{2-1}	N	H	CF	YC	GP	Grey
16	CIR 126 P_{1-2}	N	H	CF	CC	GP	White
17	CIR 126 P_{2-2}	N	H	CF	LY	GP	White
18	CIR 126 P_3	N	H	CF	CC	GP	White
19	CIR 526 P_1	N	H	CF	CC	GP	White
20	CIR 526 P_2	N	H	CR	LY	RP	White
21	CIR 526 P_3	OL	GL	YF	LY	GP	Grey
22	CIR 920 P_{1-2}	N	H	CF	CC	GP	White
23	CIR 920 P_{1-3}	N	H	CF	CC	GP	White
24	CIR 926 P_{2-1}	N	H	CF	CC	GP	White
25	CIR 926 P_{2-3}	N	H	CF	CC	GP	White

N= normal leaf; OL okra leaf; H= hairiness; GL= glabrous; CF= cream flower; YF= yellow flower; YR = yellow red; CR= cream red; LY light yellow,. RP= Red plant; GP= Green plant; RC= Red colour; YC= yellow colour; CC= cream colour.

species. Konan *et al.,* (2003) reported the introgression of genes for resistance to reniform nematode into *G. hirsutum* using *G. longicalyx* as the donor parent and *G. thurberi* as the bridging species. Badnawar 1, B 1007 and Cambodia-*tomentosum* types such as SRT 1, Khandwa 1 and Khandwa 2 are the resultant progenies from *G. hirsutum* and *G. tomentosum* crosses. These varieties are highly resistant to jassid and have velvet type of hairiness (Narayanan *et al.,* 2004).

Drought tolerance: In upland cotton, resistance to drought has been improved through genetic enhancement of germplasm lines by utilizing *desi* cottons**.** In India drought resistant breeding programmes utilizing the Asiatic cottons were also attempted by many workers which resulted in the infusing of drought resistant genes in hirsutum cotton and the release of varieties like Deviraj (170 CO2), Devitej (130 Co 2 M) and G 67 with wide adaptability (Narayanan *et al.,* 2004).Transferring drought tolerance from *Hibiscus panduraeformis* to *hirsutum* cotton is possible.

Uses of biotechnology in genetic enhancement of cotton : There are two main approaches in biotechnology which are used in pre-breeding

(i) Genetic transformation in cotton: Genetic engineering offers a directed method of pre-breeding that selectively target one or few traits for introduction into the crop plants. The development and commercial released of transgenic cotton plants that have been genetically modified relies exclusively on two basic aspects. The first being the ability to "transform" plant by introducing a gene or genes into cotton genome that are stable transmitted and express in the progeny of subsequent generation. Second gene delivery system for achieving this end is the wildly used *Agrobacterium* mediated transformation method and particle gun bombardment. The second requirement is a need to regenerate fertile plants derived from individual cells.

(ii) Cell culture and plant regeneration for genetic transformation : *In vitro* culture involves intact tissue and organs and, therefore, requires that structural integrity of the tissue is maintained. In the first instance, the objective is usually to induce the structure to develop as good as it would be on the plant. In order to achieve this, individual cells or cell clusters are cultured on nutrient media containing growth regulators. The undifferentiated mass of cells is called callus. A piece of callus is submerged in liquid medium. The cell dissociates from each other and from suspension, which can be used for somatic embryogenesis and plant regeneration. Not only have this development has enabled cotton to be genetically enhanced for desirable traits but has allowed the use of cell and organ culture for understanding basic studies on cotton genetics and physiology also.

iii) Development of transgenic cotton: The era of transgenic cotton began in 1990 with introduction of the *Bt* gene Cry1Ac to develop the first *Bt* cotton variety which showed high level of resistance to *Helicoverpa*. The gene was transferred into the genome of cotton explants using a bacterium called *Agrobacterium tumefasciens*. The transformed cells were developed into a full genetically modified plant now called Bt-cotton. The *Bt* gene from originally genetically engineered mother plant (Coker series) was transferred to advance cotton cultivars through backcrossing. In India the

Table 3. Commercial release of different *Bt* cotton events in India.

S. No	Gene (s)	Event	Developer/company	Year of Approval
1	Cry1 Ac	MON 531	Mahyco/Monsanto	2002
2	Cry1Ac and Cry2Ab	MON 15985	Mahyco/Monsanto	2006
3	Cry1Ac	Event 1	JK Agri Genetics	2006
4	Fused genes Cry1Ab and Cry1Ac	GFM Event	Nath Seeds	2006
5	Synthetic Cry1C	MLS 9124	Metahelix Life Sciences	2009
6	Cry1Ab + Cry2Ae.*	-	Bayer Crop Sciences	-
7.	Cry1Ac + Cry1F*	-	Dow Agri Sciences	-
8.	CP4 EPSPS in Roundup Ready Flex*	-	Mahyco/Monsanto	-

*Not approved for commercial cultivation

transgenic cotton was introduced into cultivation during 2002-2003 first with Bollgard I (Cry1Ac). Currently, Cry1Ac, Cry2Ab and Cry1C have been approved for commercial cultivation in India. *Bt* cotton hybrids available in India are derived from technologies developed by Monsanto (Cry1Ac and Cry1Ac + Cry2Ab), Metahelix (Cry1C), Chinese Academy of Agricultural Sciences through Nath seeds (modified Cry1Ac called as fusion gene) and JK seeds (Cry1Ac). Dow Agro Sciences are conducting field trials with Cry1Ac + Cry1F and Bayer is introducing Cry1Ab + Cry2Ae (Table 3).

The transgenic cottons were all based on proprietary germplasm and hybrids were predominantly of *G. hirsutum* x *G. hirsutum* combinations. As a result, intra *hirsutum* transgenic cotton hybrids are grown in more than 92.0 per cent of the total cotton area of 121.91 lakh hectares (Anonymous, 2014). However, Extensive cultivation of *Bt* transgenic cotton and selection pressure on target insects in a continuous mode will encourage the development of resistance in insects towards *Bt*. Therefore, *Bt* cotton will have to be managed in a way that discourages pest resistances to Bt toxins by genetic enhancement of germplasm lines through gene pyramiding. *e.g.* Monsanto and Dow, together have stacked eight genes into GM Maize called SmartStax. Six of these are for insect control, Cry1A.105, Cry2Ab2 and Cry1F for borers; Cry3Bb1, Cry34Ab1 and Cry 35Ab1 for root worms; pat for glufosinate resistance and CP4 EPSPS for glyphosate resistance. So far 342 *Bt* toxin genes are available for research to develop insect resistant genetically modified crops (Nandeshwar, 2004 and Kranti, 2012). Therefore the cotton genotypes can be genetically enhanced through genetic engineering for various characters by using different genes .

Future prospects for genetic enhancement work : In cotton distant hybridization has played a significant role in transferring the desirable characters like fineness, strength, resistance to pests like jassids and bollworm, black arm, drought resistance and male sterility besides improvement in yield by releasing hybrids. However the utilization of wild germplasm poses the various problems like reproductive barriers such as failure of pollen germination and slow pollen tube growth, elimination of chromosomes, chromosomal abnormalities, hybrid unviability and sterility (Mehetre *et al.,* 2003). Recently,

significant advances have been made in overcoming these barriers, therefore the future prospects for genetic enhancement depend on genetic engineering techniques and the wild species of cotton could be looked as a source for the contribution of genes governing the traits like fibre fineness, strength and maturity, biotic and abiotic resistance, increased photosynthetic rate, uniform maturity, etc

Though handy tools in biotechnology are available for gene introgression, it takes a long time compared to the routine hybridization programme owing to the non responding nature of cotton to the nurture given at the laboratories. Similarly, shortfalls are bound to occur in conventional hybridization owing to the disparity in the genome introgressed and genetic disturbances. Therefore there is an urgent need to encourage the Scientists to become "Genetic Enhancer or Pre Breeders" and helping in the breeding programme for development of cultivars to meet the ever increasing need of mankind on the following aspects:

- Development of genotypes suitable for early maturity with synchronous opening suitable for high density population system (HDPS) and machine picking.
- Agricultural biotechnology offers opportunity for development of germplasm lines with higher level of resistance to biotic and abiotic stresses.
- Gene transformation and marker assisted selection are useful tools to overcome the limitations of conventional breeding for the improvement of cotton germplasm lines for useful traits through interspecific hybridization or genes sourced across the species barriers.
- DNA marker assisted selection can be used in early generations to screen large numbers more efficiently.
- For developing multiple adversity resistance (MAR) lines programme in cotton collaborates actively with genome mapping, fingerprinting, gene tagging and transformation.
- Similarly to develop the germplasm lines with multiple insect resistance (MIR) to reduce the expenditure on pesticides. This can be achieved through transfer of morphological and biochemical factors imparting resistance to single genotype.
- Another concept picking up worldwide is the organic farming for which the germplasm need to be screened and genetically enhanced through pre-breeding.

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Wide hybridization in cotton – Problems and prospects

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Cotton, is one of the most important commercial crops, enjoys a pre-eminent status among all the cash crops in the country and elsewhere which has been recognized as a vital component of the global economy (Arpat *et al.,*., 2004). Cotton is a crop of prosperity having profound influence on man and matter. Currently *Gossypium* includes 50 species spreading over eight genomic groups ('A' through 'G' and 'K'), four of which are cultivated, 43 are wild diploids and three are wild tetraploids (Percival and Kohel, 1990). Out of the four cultivated species, *Gossypium hirsutum* L. and *Gossypium barbadense* L. commonly called as new world cottons are tetraploids (2n = 4x = 52), whereas *G. herbaceum* L. and *Gossypium arboreum* L. are diploids (2n = 2x = 26) and are commonly known as old world cottons. Earlier, five allopolyploid species were widely recognized, but a sixth tetraploid species *viz., G. ekmanianum* Wittm. was recently resurrected and subsequently validated (Grover *et al.,* 2015) by molecular sequence data.

Evolution of cultivated allopolyploids in different crops are first standard examples of cross between different diploid species. Species belonging to 'A' genome have been known as only fibre yielding among diploids. Two tetraploid species, *G. hirsutum* and *G. barbadense* known for cultivation were known to be evolved naturally from cross between *G. herbaceum* subsp. *africanum* (AA) and wild diploid, *G. raimondii* (DD) (Hutchinson, 1959; Douglas and Brown, 1971 and Phillips, 1974). They have been known for higher *kapas* yield as well as superior fibre quality than old world diploid cottons. *G. hirsutum* is susceptible to many biotic and abiotic stresses and genetic diversity among modern upland cotton cultivars is limited, as revealed by isozyme analysis (Wendel *et al.,* 1992) and various DNA markers including RFLP (Brubaker *et al.,* 1993; Brubaker and Wendel 1994), RAPD (Iqbal *et al., 1997*; Linos *et al.,* 2002), AFLP (Abdalla *et al.,* 2001; Iqbal *et al.,* 2001), and SSR (Lacape *et al.,* 2007, Fang *et al.,* 2013). Increasing upland cotton diversity is essential for genetic improvement. A very few of the wild species grow together in the wild, and not surprisingly therefore, documented cases of hybridization and introgression are rare (Wendel and Grover 2015). Extensive genetic variation is available among members of the genus *Gossypium* (Percival and Kohel 1990), and efforts to increase the genetic base of *G. hirsutum* can draw upon the wild species of *Gossypium*. The wild and cultivated diploid species have wide adaptability and high degree of resistance to biotic and abiotic stresses (Patel and Mehta 1990). Wild species have been utilized to transfer the resistance to various insect-pests and diseases including cotton leaf curl disease for improving the cultivated cotton species especially *G. hirsutum* (reviewed by Mehetre *et al.,* 2002c). Many reports can be found wherein researchers have been trying to produce the new polyploid species and/or introgresssing the genes from the wild to the cultivated *Gossypium* species for increasing the diversity in the modern cultivars (Ahmad *et al.,*

2011, Nazeer *at al* 2014, Zhang *et al.,* 2014, Liu *et al.,* 2015 and Mehetre *et al.,* 2009). The characterization of interspecific hybrids is another aspect of the biological research. Morphological and molecular characterization of the genotypes reveal the various genetical and epigenetical phenomena and helps in understanding inheritance of various traits as well.

Plant breeders have used unadapted germplasm, almost exclusively, as a source of major genes for insect and disease resistances and have mostly relied on repeated intercrossing of adapted elite genotypes for improvement of yield (Fulton *et al.*,, 2002).

Heterosis has been exploited in large number of crop plants. India has been a leader in the development and commercialization of cotton hybrids. The first interspecific (*G. hirsutum* x *G. barbadense*) hybrid Varalaxmi was released during 1971 (Katarki 1971). Later on, a series of hybrids *viz.,* DCH 32, HB 224, DHB 105 etc have been developed. Interspecific crosses involving *G. arboreum* x *G. herbaceum* have also been developed and released. Examples include G Cot DH 7 and DDH 2.

Interspecific hybridization of *G. hirsutum* x *G. barbadense* followed by selection in the segregating generations has led to the development of many improved cultures. Varieties MCU 2 and MCU 5 of *G. hirsutum* x *G. barbadense* cross are the successful examples. Similarly, *G. arboreum x G herbaceum, G arboreun x G anomalum, G herbaceum x G anomalum* are most exploited cases for fibre quality improvement. For example in AK 8401 (*G. arboreum*) fibre length was improved through introgression with *G. anomalum* (Kulkarni *et al.*,, 2001).

Many breeding stocks as well as varieties have been evolved so far through interspecific hybridization involving wild species. Genes for lint strength from a lintless wild species *G. thurberi* have been transferred into cotton. A triple hybrid [(*G. thurberi* Tod. × *G. arboreum* L.) × *G. hirsutum* L.] was first utilized as a genetic source for higher fibre strength (Beasley, 1940). In India, a *hirsutum* variety Arogya was developed through *hirsutum* × *anomalum* hybridization programme at CICR, Nagpur and was released for rainfed conditions in the Central Zone of India. Immunity to bacterial blight and tolerance to sucking pests were introgressed into this variety from the wild species *G. anomalum*.

Successful introgressions between *G. arboreum* and *G. hirsutum* have been reported (Deshpande *et al.*,, 1992; Kulkarni and Khadi, 1998; Deshpande and Baig, 2001). DLSA 17, an *arboreum* variety with better fibre quality has been released. Deshpande and Baig (2001) have reported the identification of segregants with increased boll weight, staple length and ginning outturn derived from interspecific crosses of *G. arboreum* and *G. hirsutum.*

Patel and Desai (1963), with an objective of improving local *G. herbaceum* types for quality characters used the Persian-211(*G. barbadense*) for crossing with improved *G. herbaceum* strains at Surat and synthetic cultures *viz.*,, 1802, 1773, 1777, 1789 and 1799 were derived from the cross (1027 ALF x Per 211) FI x 1627 A.L.F. Which possessed ginning outturn ranging from 26.1 to 40.1 per cent, fibre length ranging from 0.98" to 1.14" but, were low in yield in comparison with the local improved strains. They were therefore further crossed with the promising Surat and Buroach types to improve their yield and as a result some of the promising ones possessing good combination of yield, ginning outturn and fibre qualities were obtained. Ramachandran *et al.*,. (1964) developed hybrids involving *G. anomalum* with *G. arboreum* cultures like, 5001,

6874 and B-32-48. The results were assessed in the first and second back crosses and also in the straight crosses of the above hybrids in the advanced generations. The composite samples of lint of selected families showed the fibre weight to be 0.213 millionth of an ounce per inch compared to 0.200 for *G. arboreum*, a pressely strength index of 8.94 lb/mgm compared to 7.8 lb/mgm for *G. arboreum*. In the progeny of the hybrid BC_2F_2 of 6874 x *G. anomalum,* a pressley strength index of as high as 9.82 lb/mgm and fineness of 0.093 millionth of an ounce per inch were recorded.

Katarki (1971) carried out the interspecific hybridization between *G. hirsutum* x *G. barbadense* and evolved the first commercial interspecific hybrid, Varalaxmi. It showed superior fibre quality characters like fibre strength (44.8 at 'o' guage), fibre fineness (3.2 micronaire) and fibre length (32.7 mm). Later he released another high yielding interspecific hybrid DCH-32 which was superior to Varalaxmi in yield potential and ginning out turn. Hyer (1973) derived nearly isogenic lines from the sixth back-cross generation of 41- 63 x Acala 4-42-77 back-crossed to Acala 4-42-77. He reported that the glanded and glandless lines were similar in fibre properties but the lint yield of the glandless line was lower than that of the glanded line. The genes gl2 and gl3 (both for glandlessness), or genes closely linked to these therefore appear to depress lint yield without affecting fibre properties.

Meyer (1974) transferred characters such as nectariless from *G. tomentosum*, fibrous root character from *G. sturti*, increased fibre strength from *G. thurberi*, bollworm resistance from *G. anomalum* and cytoplasm of *G. tomentosum*, for development of male sterile hybrids into cultivated tetraploid, *G. hirsutum*. Nectarilessness is a necessary character to prevent outcrossing and impart pest tolerance. Meyer and Meredith (1978) were able to transfer nectarilessness character from *G. tomentosum* to upland cotton and cultures were popularised as DESTOM16. *G. herknessii, G. aridum* and *G. trilobum* have been used as donors for cytoplasmic-genetic male sterility system. However, due to yield penality, these systems have not been commercialized so far.

Niu *et al.,* (1998) used two cultivated species (*G. hirsutum* and *G. arboreum*), four wild species (*G. thurberi, G. anomalum*, *G. sturtianum* and *G. bickii*) and one semi-wild species (*G. mexicanum*) to create 76 new germplasm lines of nine types through various methods of hybridization, *in vitro* culture, selection, identification and multiplication. These lines have various desirable characteristics including high fibre quality (specific strength and fineness higher than those of the current popular varieties). They reported that some lines (BZ701-712) had a span length of 33.4-37.7 mm and some lines (BZ901-903) were very early having a growth period of 100-115 days.

Katageri *et al.,* (2004) reported the results of interspecific hybridization using *G. barbadense* as donor and *G. herbaceum* and *G. arboreum* as recipient parents. Selected recipient plants of Jayadhar possessed fibre lengh of 24 – 26 mm, fibre strength of 20 – 23 g/tex as against 22 mm and 16 g/tex of Jayadhar. Similarly, selected A-82-1 *arboreum* plants had fibre length 24 mm and 25 g/t against 16 mm and 13g/t of the recipient. Plants with 24 g per tex fibre strength and 28 mm fibre length were isolated in F_3 from cross between *G. hirsutum* var. Abadhita x (G.cot- 11 x *G. tomentosum*) by Soregaon (2004).

Ahton *et al.,* (2003) investigated mating schemes to achieve *G. sturtianum* and *G. australe* diploid cottons into tetraploid *G. hirsutum*. They

were able to obtain seven different single chromosomes of *australe* in *hirsutum* background. These lines constitute valuable materials for carrying out fundamental and applied genetic investigations.

Pima cottons possess superior fibre properties than the upland cottons. Saha *et al.,* (2004) reported the development of substitution lines in the *hirsutum* background where a chromosome/chromosome arm of *hirsutum* genome was substituted by a corresponding chromosome/chromosome arm of *barbadense* genome. The lines with substitutions for chromosomes 15, 18, 14sh and 22sh had reduced seed cotton yield and lint yield. Lines with alien chromosomes 2, 6, 16, 18, 5sh, 22Lo, and 22sh had improved lint percentage. Lines with substitution of chromosome 25 had reduced micronaire and increased fibre length. All the substituted chromosomes except 2,4 and 6 had reduced boll weight. Lines with substituted 14sh, 15sh and 25 had increased fibre length. The results provided information on the association of specific chromosomes with genes agronomic and fibre traits. These new genomic resources will, provide additional approaches for improvement of upland cotton and will enable the development of chromosome specific recombinant inbred lines for higher resolution mapping.

Bacterial blight resistance from *G. anomalum* to *G. hirsutum* in the form of short duration short staple drought tolerant variety Arogya which was released for cultivation in the Vidharba region (Gotmare *et al.,*, 2004).

Besides lint, cottonseed is also an important source of edible oil. Cottonseed oil is composed of three fatty acids *viz.,*, linoleic acid, olic acid and palmitic acid and two minor fatty acid namely myristic and stearic acids. Gossypol has to be removed from the oil that increases the processing cost. Gossypol is considered to impart resistance/tolerance to some insect pests. Therefore, it would be desirable to have plants with gossypol present in the shoot but absent in the seeds. Vroh *et al.,* (1999) synthesized two trispecies hybrids *G. thurberi - G.sturtianum - G.hirutum and G. hirsutum – G. raimndii – G. sturtianum.* Backcrosses with the *hirsutum* parent were made. The gland levels in the backcrossed seeds ranged from glandless seeds to normally glanded seeds. All vegetative parts of those hybrids were glanded, but a wide range of variability for gland density was observed on leaf stem, bract and calyx. Plants derived from seeds having a reduced level of gossypol constitute very interesting germplasm to develop cultivated glanded cotton with low gossypol seeds.

Mehetre (2010) introgressed successfully the desired traits like fineness and strength, non-convoluted and fully thickened with secondary cellulose deposition and thinnest fibre from wild *Gossypium anomalum* into cultivated varieties blending both conventional and nonconventional techniques. The recovered transgressive segregants would afford vast opportunities in selecting plants with desired attributes from segregating populations and introgression of genes from wild *Gossypium anomalum* to commercial cultivars.

Another study attempted to explore the possibility of successfully transferring the jassid resistant genes from two wild cotton species *G. armourianum* and *G. raimondii* into the cultivated *G. hirsutum* genotypes through backcrossing and colchiploidy (Pushpam and Raveendran 2006). The percentage of boll set was maximum in the cross *G. hirsutum* x *G. raimondii* (14.3%) and minimum in the cross *G. hirsutum* x *G. armourianum* (7.5 %). Viable seeds were obtained in all combinations indicating the compatibility between all the *hirsutum* lines with the wild

species *G. armourianum* and *G. raimondii*. The hybrids between *G. hirsutum* x *G. armourianum* and *G. hirsutum* x *G. raimondii* involving wild species as seed parents were highly sterile. Also there was no boll set on self-pollination as well as backcrossing with the cultivated parent. Although about 3400 crosses were effected, no boll setting was observed. Nevertheless, in reciprocal backcrosses when the F_1 was used as pollen parent on *G. hirsutum* female parents, a few bolls were obtained.

Sacks and Robinson (2009) introgressed resistance to *Rotylenchulus reniformis* into the cultivated tetraploid species *G. hirsutum* through crossing a resistant diploid A_2-genome *G. arboreum* with a hexaploid *G. hirsutum*/ *G. aridum* bridging line $2[(AD)_1D_4]$ to obtain a tetraploid triple species hybrid.

Nazeer *et al.,* (2014a) explored the possibility of transferring virus resistant genes from the wild species *G. stocksii* into *G. hirsutum*. Interspecific F_1 hybrid between the two species was produced after attempting 438 pollinations. 3.4 per cent boll setting and 42.9 per cent germination was obtained during this hybridization programme. F_1 seeds were treated with 0.03 per cent colchicine to obtain hexaploid plants with the success rate of 33.3 per cent. The F_1 population did not show any symptom of Cotton leaf curl disease (CLCuD) in the field, tested by grafting with CLCuD susceptible rootstock. It was concluded that it is possible to transfer CLCuD resistance and high fiber strength from *G. stocksii* to *G. hirsutum*.

A new synthetic allotetraploid ($A_1A_1G_2G_2$) between *G. herbaceum* and *G. australe* was produced by Liu *et al.,* (2015) to transfer "Glandless-seed and Glanded plant" trait to the upland cotton in a period of 9 years. A total of 16 putative hybrid seeds were obtained from 200 pollinated flowers and planted in 2007. At the maturity stage, only one plant resembled *G. australe*, which was a putative F_1 interspecific hybrid ($2n = 2x = A_1G_2 = 26$). The putative interspecific F_1 hybrid plant appeared to be highly male and female sterile, as no pollen was released and no bolls were produced when the plant was pollinated by *G. herbaceum*. The sole putative hybrid plant was propagated by grafting and treated with 0.10% colchicine for 24 h during squaring stage. In the sixth year, one branch of the hybrid plant had produced three bolls and a total of 19 S_1 seeds were obtained from these bolls by self-pollination in 2012. This interspecific tetraploid hybrid had partial fertility. In 2013, S_1 seeds (derived from the grafted hybrid F_1) were planted on soil in small plastic pots. One S_1 plant was rescued which set five bolls to give 22 seeds, while the other two S_1 seedlings. In 2014, S_2 seeds were planted. The interspecific incompatibility, to some extent, had been alleviated in the S_2 generation. Both S_1 and S_2 were new synthetic allotetraploid plants.

According to Anjum *et al.,* (2015), there is no resistant genotype in upland cotton to CLCuD, the only way is to introgress this resistance in upland cotton from wild species. 30 *Gossypium* species were screened for resistance to CLCuD and ten diploid species were found to be resistant to Burewala strain of cotton leaf curl virus. They reported that material with good fibre quality traits and resistance to various insects and pests is being developed using wild diploid species.

A project is underway across many centres in the country for the enhancement of fibre quality in *desi* cottons. As a result, many cultures with superior fibre properties and acceptable yield level have been developed.

Cotton leaf curl disease (CLCuD) is a serious threat to American cotton cultivation in the north Indian cotton growing states of Punjab,

Haryana and Rajasthan. Heavy yield losses due to CLCuD have been reported especially if the disease appears at an early stage of crop growth. *G. arboreum* possesses high degree of resistance to this dreaded disease and can serve as an important donor for the transfer of CLCuD resistance into American cotton. At PAU, crosses between *G. hirsutum* (AADD) x *G. armourianum* (DD) have been attempted and efforts are underway to induce amphidiploidy in the resulting sterile triploid hybrid.

Fertilization barriers in interspecific crosses and subsequent generations : A major objective in most of breeding programs is to generate the genetic variability for improving economic traits of crop. Hybridization among species and between species generates considerable amount of genetic variation, which could be used for further selection of desirable traits. But, several pre and post-zygotic barriers prevent the successful gene transfer from wild to cultivated species reviewed by Mehetre *et al.,* (2002a).

Failure of pollen germination is an important incompatibility barrier in obtaining wide crosses. Peng and Qian (1989) observed that the triploid F_1 pollen grains from the cross between *G. hirsutum* x *G. raimondii* were not germinated on the stigma or showed partial germination and abnormal growth. Slow pollen tube growth is one of the major crossability barriers restricting fertilization in wide crosses of cotton. Govilla (1970) found that pollen tube of *G. arboretum* pollen fails to reach ovules of *G. raimondii*, which has longer style than *G. arboretum.* Baloch *et al.,* (2000) observed that difference in style length of tetraploid and diploid species was significant, with tetraploid species having two times longer style than that of diploid species. So, they predicted that differential reproductive structures could at least be partially responsible for crossing failure and reciprocal crosses may be tried for successful fertilization. Study by Saravanan *et al.,* (2010) also supports the work of Baloch *et al.,* (2000) as they reported severe reduction in number of pollen tubes as they grew in style depending on initial pollen load. Apart from arrest of the pollen tube at different levels, several abnormalities like twisting and bulging of the pollen tube, knot formation in pollen tube, lateral enlargement of pollen tube and growth of pollen tube in opposite direction were noticed. Sometimes contents of the pollen tube are not released in the ovule**.** Shakhmedova (1981) reported that pollen tube did not enter the ovules in the cross of *G. hirsutum* x *G. anomalum.*

Post fertilization barriers hinder or retard the development of the zygote after fertilization and normal development of the seed. They include reproductive abnormalities in F_1 hybrids and their later generation progenies. Hybrid inviabilty or weakness may be due to several mechanisms affecting the development of the zygote from the first cell division after fertilization and up to the final differentiation of reproductive organs and formation of gametes. Weaver (1957, 1958) carried out embryological studies in *G. hirsutum* x *G. arboreum* direct as well as reciprocal crosses. Weaver attributed incompatibility in direct cross to physiological imbalance between hybrid embryo and hybrid endosperm. This imbalance caused cessation of embryo development 6 days after pollination (DAP) leading to embryo starvation. In reciprocal crosses, many of nuclei start abnormal divisions giving rise to structural abnormalities like dumb-bell shaped nuclei, large vacuoles and clumping of nuclei.

As a result of wide hybridization, there are two different types of genomes present in a

nucleus, due to this, hybrid sterility may arise because of differences in structure and number of chromosomes, lack of chromosome homology resulting in variable number of univalents and production of unbalanced gametes. When we cross tetraploid species with diploid species of *Gossypium* triploid F_1 so form is expected to be sterile because of production of unbalanced gametes. This is common in case of crosses between new and old world cottons *G. hirsutum* x *G. arboreum*. (Gill and Bajaj 1987; Mehetre *et al.,* 2007; Nazeer *et al.,* 2014b), *G. arboreum* x *G. anomalum* (Mehetre *et al.,* 2004b), *G. hirsutum* x *G. raimondii* (Saravanan *et al.,* 2007) etc. Hybrid breakdown is a type of reproductive failure defined as inviability or sterility observed only in the F_2 or later generations of interspecific crosses, while F_1 hybrids are viable and fully fertile. Even if hybrid have been produced successfully and are found to fertile, hybrid breakdown is the next problem it may encounter.

Overcoming cross compatibiltiy : Incompatability observed in wide hybridization of crop plants is a major hurdle in introgression of genes from wild to cultivated species. Boll setting in interspecific crosses of *Gossypium* is limited by physiological boll drop and boll shedding due to injury to pistil during emasculation and pollination. There are reports of increase in boll retention after application of growth regulators. Gill and Bajaj (1987) used mixture of 50 ppm GA_3 + 100 ppm NAA for increasing boll retention in *G. hirsutum* x *G. arboreum* and reciprocal crosses. They applied mixture of growth regulators at base of pedicle for three consecutive days starting from one day after pollination. They reported that boll retention varied between 52.9 - 79 per cent in *G. hirsutum* x *G. arboreum* and 43.7- 60.7 per cent in *G. arboreum* x *G. hirsutum* after application of growth regulator as compared to 6.4- 17.6 per cent retention in the control, where no growth regulator has been applied.

Altmann (1988) observed that cotyledons appeared more fully developed when NOA/GA and NAA/GA treatment were used. Further, reduction in boll absicission with 50 ppm GA_3 + 100 ppm NAA was recorded along with increase in mean boll weight. Also, hormonal treatment resulted in more embryos per boll and improvement of embryo quality. He concluded that application of optimum levels of growth regulators was superior to embryo/ovule culture in obtaining interspecific hybrids in most of the crosses performed during this investigation.

In an another investigation, efforts were made by Mehetre *et al.,* (2002b) to induce boll setting in interspecific hybrids in which boll and seed setting was a problem, so as to use the ovules from these bolls to supplement *in-ovulo* embryo culture. Hence to exploit these hybrids for the introgression of desirable genes from wild to cultivated cotton 16 different treatments, i.e. combination of (i) without sugar and agar, (ii) sugar 30 per cent, (iii) sugar 30 per cent and agar 0.04 per cent with (a) GA_3 50 ppm + NAA 40 ppm, (b) GA_3 50 ppm + NAA 100 ppm, (c) NAA 100 ppm and (d) IAA 100 ppm were used. Out of all tried treatment containing mixed pollen in 30 per cent sugar + GA_3 50 ppm + NAA 100 ppm was found to be effective in inducing boll setting on interspecific hybrids. Also it was observed that different treatments gave different results on different genetic background.

Usefulness of growth regulators in improving crossed boll retention was investigated by Pathak *et al.,* (2003) in intra-*hirsutum* crosses. In general, they observed increased retention of crossed bolls after application of 50 ppm GA_3 + 100 ppm NAA. Only one *G. hirsutum* genotype May Acala gave an

overall negative response to hormonal treatment. So, it was suggested that response of genotypes to growth regulators depends on endogeneous hormonal levels. Similar study was conducted in inter varietal crosses of *G. arboreum* (Pathak *et al.,* 2008), where significant differences between mean value of boll retention of direct (15.12%) and reciprocal (24.99 %) crosses was observed.

Rauf *et al.,* (2006) crossed *G. hirsutum* and colchicoid *G. arboreum* in both direct and reciprocal manner. Direct crosses proved successful but reciprocal crosses were complete failure so they tested different treatments of hormonal application for inducing boll setting in reciprocal crosses. They observed highest number of interspecific bolls or seeds per boll when 0.5 mg/l GA_3 and 0.5 mg/l IAA was used in *G. arboreum* genotype FDH-228 and HK-113 respectively. They also reported genotypic specificity to different hormonal treatments. Growth regulators have been successfully used to develop interspecific hybrids in other crops as well *e.g.,* chickpea (Verma *et al.,* 1990), pigeonpea (Sidhu *et al.,* 2000), soybean (Singh *et al.,* 1990) etc.

Even though there are reports of development of interspecific hybrids in cotton through conventional breeding approaches but success is limited due to various barriers, so tissue culture techniques offers an alternative for obtaining interspecific hybrids. One biotechnological technique for overcoming the post fertilization barriers is embryo culture. The first report on successful *in vitro* embryo culture was by Hanning (1904) who grew nearly mature embryos of *Raphanus* and *Cochleria* on mineral salts medium with sugars, amino acids and plant extracts. Laibach (1925) was first to successfully culture hybrid embryos from interspecific crosses of genus *Linum* which normally resulted in aborted seeds.

The first use of embryo culture in cotton was reported by Skovsted (1935). A weak embryo of *G. davidsonii* x *G. sturtianum* was rescued and cultured on sterile glucose-agar media. Beasley (1940) cultured interspecific *Gossypium* species hybrids on White's media. Some of them germinated and formed roots and hypocotyls but did not develop further. He also performed embryological studies to determine the cause of incompatibility and observed that degeneration of endosperm lead to embryo starvation in both *G. hirsutum* x *G. arboreum* and reciprocal cross. Lofland (1950), Dure and Jensen (1957) were successful in obtaining seedlings when embryo was excised 20-25 days post anthesis. However, younger embryos failed to mature *in vitro*.

In vitro culturing of *G. hirsutum* embryos from heart stage to maturity by adjusting osmotic potential of media and using high salt media was reported by Mauney (1961). In a later study, Mauney *et al.,* (1967) performed chromatographic analysis of liquid endosperm from 12-14 day old cotton ovules and found that malic acid (7 mg/ml) was major organic acid and further reported that addition of calcium or ammonium malate (upto 4 mg/ml) to media improved growth and viability of cotton embryos cultured at heart stage. Malate was also observed to effect osmotic pressure of medium. So, embryo could be cultured for longer period of time on low osmotic pressure medium. This eliminated the need for addition of NaCl to maintain osmotic pressure at 10 atm as suggested by Mauney (1961).

After this, the focus of researchers shifted to *in ovulo* embryo culture as it proved to be more simpler and successful method for rescuing embryos. Culturing of ovules is advantageous because these can be easily excised at zygote stage and also provides a "maternal environment" to growing embryo.

Joshi (1960) first reported ovule culture in *Gossypium*. Six DAP ovules were excised and cultured on low salt medium containing casein hydrolysate, vitamins, indoleacetic acid (IAA) and gibberellic acid (GA). Growth of ovules was abnormal and fibres did not grow. This culture method was later modified and ovule growth response was documented by Joshi and Johri (1972). They studied the effect of IAA, KN, GA_3, CH and YE on *in vitro* growth of selfed cotton ovules excised 6 DAP. They observed embryo growth upto early dicotyledonous stage on White's medium containing higher concentration (1000-2000 ppm) of CH, while on lower concentration (upto 250 ppm), embryo grew only upto globular stage. Also, higher concentration of IAA (1-2.5 ppm) did not favour embryo development. Fully differentiated embryos were obtained 96 days after fetilization from 12-celled pro-embryo.

The first successful method of ovule culture for fiber development was given by Beasley *et al.,* (1971). The essential features for success were the use of high salt *Bt* media and use of liquid culture instead of agar solidified medium. However, this report was only concerned with development of ovules and fibres for two weeks and no embryos developed to maturity. Beasley and Ting (1973) studied effect of phyto-hormones (BTP medium) on fiber development in fertilized and unfertilized ovules. They reported that fertilized ovules (2DAP) did not require hormones and exogeneous IAA did not promote additional fibre growth. Addition of exogeneous GA_3, however, had stimulatory effect, while, kinetin and abscisic acid (ABA) were observed to be inhibitory. In case of unfertilized ovules IAA and GA_3 had additive affect for fibre growth. Exogeneous kinetin promoted increase in ovule size but did not support fibre development. They further stated that disposition of parent plant greatly affected the capacity of ovules to grow *in vitro* and also their response to applied growth substances. At the same time, a Belgium group (DeLange and Eid 1971, Eid *et al.,* 1973) published report concerning fibre growth on cultured ovules and found that MS was superior to media with lower salt formulations. They concluded that relatively high nitrogen content of MS was advantageous. They examined the effect of auxin and GA_3 and reached same conclusion as Beasley and Ting (1973).

Stewart and Hsu (1977) found that BTP medium (*Bt* medium with phytohormones) was basically correct for *in ovulo* embryo growth except for poor development of cotyledons. They noted that supplementation of BTP medium with 10-15 mM ammonium ions supported ovule growth and germination of embryos. Further, they observed that after germination, radical tip became necrotic and no lateral roots were formed. Subsequent observations indicated that seedlings were unable to tolerate high-salt or high-sugar media and inositol was inhibitory to root development. Low salt media without inositol allowed balanced root and shoot growth with two to three leaves. They also reported differences in germination response with respect to cultivars. In subsequent study, Stewart and Hsu (1978) reported culturing of 2-4 DAP ovules to obtain interspecific hybrids between Asiatic diploid and American tetraploid cottons in all possible combinations. In most cases, the presence of GA and kinetin was deleterious to recovery of hybrid plants. Consequently, they recommended that only auxin be used when culturing ovules of species other than *G. hirsutum*.

In a similar study, Umbeck and Stewart (1985) recovered interspecific hybrids from eight of twelve crosses made using three wild species

as maternal parent and four cultivated species of cotton as paternal parent. They applied GA_3 (3.5 mmol/l) to the flower at anthesis and immature embryos were rescued 15 – 25 DAP and cultured on medium of Stewart and Hsu (1977). They observed that success of fertilization and embryo development was strongly influenced by paternal parent used and degree of hybrid embryo development might be a more important factor than age or size at the time of embryo rescue.

Interspecific hybrids between cultivated diploid species and wild diploid species were obtained by Gill and Bajaj (1984) using MS media supplemented with IAA (1.5 mg/l), kinetin (0.5mg/l) and casein hydrolysate (250mg/l). They applied mixture of growth regulators (NAA @ 100mg/l + GA_3@ 50mg/l) at base of pedicel for 5-8 DAP to prevent early boll shedding. Later on, Gill and Bajaj (1987) used same method to obtain hybrids between *G. arboreum* x *G. hirsutum* by culturing 3 days old ovules. Similar work was done by Kalamani (1996). However, Thengane *et al.,* (1986) observed that no single medium was adequate to ensure complete development of fertilized ovules, hence, they performed sequential five step transfer to different media to obtain interspecific hybrid between *G. hirsutum* cv. Laxmi and *G. arboreum* cv. Jyoti. Dhumale *et al.,* (1996) used BT and MS media for obtaining hybrids in three varieties of *G. arboreum* and four varieties of *G. hirsutum* in different combinations and found that response of hybrid embryos to media was genotype oriented. *In ovulo* embryo culture was used to rescue hybrid between amphidiploid (*G. arboreum* x *G. anomalum*) and *G. hirsutum* by Mehetre *et al.,* (2004b) for combining desirable traits of diploids into cultivated tetraploid.

To improve effectiveness of obtaining interspecific hybrids of *Gossypium* species, Sacks (2008) compared nine media modifications based on earlier studies. He observed highest frequency of germination for MS media fortified with Gamborg's B5 vitamins and additional 1.9g/l KNO_3. They also observed that media without phytohormones produced more seedlings than the media in which different combinations of IAA and kinetin were tested. This was contrary to results of Gill and Bajaj (1987) who observed no germination of *G. hirsutum* x *G. arboreum* without growth regulators. However, Sacks pointed out that differential genotypes used in two studies might account for differing results obtained by addition of auxin and kinetin.

Quantification of level of major and minor elements, carbohydrates, ammonium ions, free amino acid and hormones in cotton ovules, nucelli, and ovule fluid was done by Fuller *et al.,* (2009) to develop tissue culture media that normalized development of early stage globular embryos *in vitro*. To further extend this work, Liddiard and Carman (2010) quantified dissolved oxygen tensions, osmotic potentials, and pH at several locations in cotton ovules during embryony and also reported the procedures to normalize early development of embryos *in vitro* by simulating these parameters. Based on these analyses, Fuller and co-workers (2011) devised chemically defined media and unique culture equipments for inducing rapid differentiation, growth, and germination of cotton embryos starting with globular pro-embryo as explants. They recommended GOS1 formulation for culture of cotton pro-embryos with addition of IAA, mannitol, and phytagel. This formulation differed from MS media by containing 8.2-fold more P, 2.4-fold more K, 2.7-fold more Ca, 5.1-fold more S, 8.8-fold more myoinositol, and arabinose, melibiose, malic acid, citric acid, a variety of amino acids, and mannitol (as an osmoticum, "1.10 Mpa), none of which are in MS

medium.

Although most of the researchers have reported the successful production of interspecific hybrids and their further generations in *Gossypium* genus using embryo rescue technique, few have reported failure as well (Altman *et al.,* 1987). So, one can use both the techniques- embryo rescue and direct crosses in field, simultaneously.

Research utilizing *Gossypium* germplasm is essential, because this is a complex genus. Although much has been accomplished in the understanding of the genus, much remains to be done This genus comprises species with differing ploidy levels and represents a high degree of variability, from highly improved allotetraploid species to wild diploid species. This variability has only begun to be tapped as a source of beneficial characteristics.

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Plant ideotype breeding in cotton

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Abstract : Globally India ranks first in respect of area under cotton cultivation and production. However the productivity remains stagnant for the past few years and some innovative approaches are the need of the hour to break the yield barrier. Conventionally, cotton is cultivated with wider spacing and hence breeders concentrate on increasing the per plant productivity by developing robust plant types. Based on recent experiences in various demonstrations, it has been clearly shown that the yield level of cotton can be enhanced by growing semi-compact cotton genotypes under high density planting system. Cotton varieties with ideal plant type (ideotype) are to be developed to suit the growing conditions in HDPS. The concept of ideotype breeding in cotton is discussed.

Keywords: Cotton, ideotype, plant architecture, super okra leaf type

With an ever increasing human population and gravity of both biotic and abiotic stress, there is a great necessity to breed crop cultivar for adaptation under these stress situations as well as with good high yield potentials to ensure global food security. The yield potential is defined as the yield of a variety when grown in fluctuating and adverse situations against biotic and abiotic stresses effectively controlled. Stress factors such as drought and salinity offer a great challenge to the breeders to breed crop cultivars tolerant to the gravity of these stress factors as alternative to assure food security. The identification of ideotype for a particular crop could be of great advantage to the plant breeders.

Cotton area increased steadily in India with the introduction of transgenic cotton in 2002-2003 and presently over 90 per cent of the cotton grown is represented by *Bt* cotton hybrids belonging mostly to *G. hirsutum*. During the last few years, the cotton production has increased considerably and thereby surpassing China to reach highest producer of cotton (Anonymous, 2015). This is mainly because of increased acreage under cotton cultivation in India. In India, more than two third of area under cotton cultivation is rainfed. The cotton productivity remains a major concern, which is stagnated at around 500 kg lint/ha during the last few years.

The *Bt* cotton hybrids are being cultivated at a wider spacing of either 120 x 90 or 90 cm x 90 cm or even wider at some places with a plant population of 9,000 to 12,000 plants/ha even under rainfed situations. *Bt* cotton hybrid seems to be a suitable option only for irrigated areas with high input management (Manickam *et al.*, 2014b). However, under rainfed situation with marginal soils, the yield levels of *Bt* cotton hybrid remains to be very low.

Low cotton prices, increasing production costs, and vagaries of monsoon are seriously challenging the cotton farmers in the country. Promising strategy for overcoming this economic challenge is to identify production practices that increase yield or reduce cost. From a practical standpoint, identifying and implementing production practices that increase yield or

reduce costs within the conventional cotton production system will be difficult. In contrast, High Density Planting System (HDPS) cotton production is of commercial interest because of its potential to reduce costs while still producing acceptable fiber quality and yield. However, these systems will be successful only if we develop varieties with suitable morphological attributes (ideotype) for realizing higher yield.

The concepts of building ideotype model is based on the morphological and reproductive traits associated with efficient capture of sunlight for photosynthesis and translocation of the photosynthates to the sink (economic product). Development of the ideotype concept has focused the attention of plant physiologists on identification of simple morphological characters which have some influence on physiological processes determining the yield of the economic organs (Thurling, 1991). Characters such as leaf inclination and leaf shape, for example, are often simply inherited and can greatly influence crop canopy structure and radiation interception. Such characters could be rapidly modified by selection to increase crop photosynthesis and yield. Ideotype definitions integrate information on these relationships and provide plant breeders with a clear blueprint of the characteristics of a high-yielding cultivar in a specified environment (Thurling, 1991).

Plant breeders have attempted to enhance yield by selecting for individual traits since the beginning of plant breeding. This approach has been broadened to encompass the breeding of model plants or ideotypes. An ideotype is a hypothetical plant described in terms of traits that are thought to enhance genetic yield potential. Ideotype breeding is defined as a method of breeding to enhance genetic yield potential based on modifying individual traits where the breeding goal (phenotype) for each trait is specified. Successes have been reported in breeding to enhance yield with individual traits, the value of genetic diversity for individual traits, and benefits from goal setting which support ideotype breeding. Requirement of symmetry in size among plant parts, compensation among plant parts, pleiotropy, and genetic background may slow progress in ideotype breeding. Ideotype breeding is recommended as a methodology to augment traditional plant breeding, when the breeding goal is enhancing genetic yield potential. Breeding experience and research to date suggest that ideotype breeding is not a suitable substitute for traditional yield breeding (Rasmusson, 1987).

Studies of genetic architecture, combined with improved knowledge of the structure of plant population, will impact the understanding of plant evolution and the design of crop improvement strategies to enhance plant growth, development and productivity (Holland, 2007). Higher plants display a variety of architectures that are defined by the degree of branching, internodal elongation, and shoot determinancy (Wang and Li, 2008). Crop yields depend on a canopy's capacity to intercept and efficiently use solar radiation. Photosynthetically Active Radiation (PAR) represents the solar radiation that can be absorbed by green plants and used for photosynthesis to produce biomass. Canopy architectural information is essential to a mechanistic description of radiation interception.

The architecture of a plant depends on the nature and relative arrangement of each of its parts; it is, at any given time, the expression of an equilibrium between endogenous growth processes and exogenous constraints exerted by the environment (Barthélémy and Caraglio,

2007). The endogenous regulatory principles that control plant architecture were documented by Reinhardt and Kuhlemeier (2002). They propose that plant architecture is species specific, indicating that it is under strict genetic control, although it is also influenced by environmental conditions such as light, temperature, humidity and nutrient status. In addition, the basis of leaf architecture and the role of cell division and cell growth in morphogenesis influence plant architecture.

In cotton, the ideal ideotype under high input situation should be open leaf canopy, long stout and strong petiole for efficient interception of light energy; stout strong stem, intermediate internode length, big boll size, high boll number, sympodial branches of medium length to accommodate more plants between rows and strong deep rooted (Maiti and Rodriguez, 2010). Plant architecture determines the productive potential of a crop species. Plants with crowded leaves indicate wastage of resources, light energy, water and nutrients especially under drought situations. Heitholt *et al.,* (1996) indicated that a leaf shape variant with reduced leaf area (okra leaf) yielded more in 51 cm rows than in 102 cm rows. The 76 cm row spacing intercepted significantly more percentage of photosynthetic photon flux density than 102 cm rows up until 80 d after planting. Cotton in 76 cm rows outyielded those in 102 cm rows by 15 per cent.

Manickam *et al.,* (2014a) demonstrated that the yield potential of cotton can be improved by growing superokra leaf type plants having sucking pest tolerance cultivated at a closer spacing of 45 x 15 cm accommodating around 1,50,000 plant/ha. It has been indicated that such genotypes are amenable for further closer spacing of 37.5 x 10 cm, thus accommodating more than 2,50,000 plants/ha.

According to Chunsong (1993) ideotype cultivation of cotton refers to a cultivation system aiming at producing maximum fine quality bolls by choosing suitable varieties, date of planting, planting pattern so as to have a long good quality boll formation period. When the required total sympodia are reached, chemical control are to be employed to minimize the underfed vegetative growth, and to establish the maximum fine quality bolls to obtain top quality and high yield through the optimal combination of boll number and boll weight. Further he indicated that that the daily increment of plant height of cotton is a sensitive trait which may affect the growth and development processes of cotton plant.

Early varieties with a compact habit could adapt to a shorter rainfall cycle (Constable, 1998) and that their low spatial footprint would enable these plants to withstand higher stand densities, thus compensating for their lower individual yields.

Sekiloka *et al.,* (2008) studied production and development patterns in 10 cotton varieties to determine the most efficient strategies that could be transformed into breeding traits. They identified two ideotypes that yielded better than the others *viz.,* for late planting and high stand density conditions, low vegetative growth, early flowering onset, a short flowering period and low boll retention at the first position of the fruiting branches were desirable, whereas, for early planting and low stand density conditions, high vegetative growth, late flowering, long effective flowering time and high boll retention at the first position of the fruiting branches were desirable.

Planting date and density differentiated growth and development greatly (Galanopoulou-Sendouka *et al.*, 1980). The influence of planting date was strongest on earliness, while density affected most morphological characteristics and

yield components. Increasing density reduced individual plant growth and productivity. However, in per unit land area, there was higher total dry matter production, earlier and more abundant foliage and fruiting, but not a proportional increase of economic yield and earliness. The very dense plants showed lower efficiency of leaves, higher fruit shedding, and smaller boll size. Medium density compared to high density was more consistent in earliness and high yield.

There is strong interest to develop cotton cultivars that reduce the time from planting to maturity which can lead to savings in irrigation water and late insect pest season spray costs. However, there is generally a trade off in seeking earlier maturity (*i.e.* shortening the time between planting and harvest), in that for each day that maturity is brought forward, there is a yield loss of between 20 and 35 kg/ha/day, that is between 0.6 to 1.0 bales/ha/week (Bange and Milroy 2004). It is generally understood that the timing of crop maturity in cotton is not determined solely by temperature and day length as in many other crops, but by the balance of supply and demand of resources to the developing fruit and growing points.

The Ultra Narrow Row (UNR) plants were shorter, had fewer nodes, and had fewer total sympodia than the conventional plants. The UNR plants had fewer bolls than conventional plants, with a higher percentage of the total boll number in the first sympodial position and a lower percentage in the second position. Higher seed cotton yield of UNR cotton appeared to result from the higher plant populations (Vories and Glover, 2006). While there is no standard definition, the term "ultra narrow row" cotton has generally referred to cotton grown on a row spacing of 38 cm or less without post plant cultivation and harvested with a stripper. The UNR plants were consistently shorter than conventional plants, an average of 17 cm less, which is an advantage for stripper harvesting. The UNR plants averaged 3.4 fewer nodes than conventional; however, even with the reduction in numbers of nodes, the average internode length averaged 4 mm less for UNR. Similarly, significantly higher yields of cotton were obtained for UNR when compared to conventional rows (Wiatrak *et al.*, 1998). However, the cost required to produce UNR and conventional cotton was similar (Nelson *et al.*, 2001).

In Indian conditions, Patil *et al.,* (2007) compared robust and compact cotton cultivars in an experiment in which the two groups of plant types were grown in different plant spacings depending on their plant morphology. They confirmed that compact cotton phenotypes can produce high yields/unit area of production. Analysis of the path to productivity of these genotypes indicated that robust types occupied greater three dimensional space by virtue of greater plant height, longer monopodia and sympodial length, and greater sympodial internode length. Compact types, conversely, had shorter main culms and sympodial internode length. Even though the/sympodia and per plant boll numbers and boll weight were lower in compact phenotypes, boll number/unit area was higher. The best compact phenotype produced bolls with the most efficient use of its three dimensional space .The contrasting strengths of robust and compact types suggest a need for developing an intermediate plant type that blends the features of higher three dimensional space but lesser horizontal growth to improve seed cotton yield.

Improvements in carbohydrate source–sink relations are needed to improve efficiency of yield formation in cotton (*Gossypium hirsutum* L.). Most source sink research has focused on

leaf boll relationships, with little study of vegetative storage reserves. Possible ways of altering the source–sink balance include increasing the Plant Population Density (PPD) and using a plant growth regulator, Mepiquat Chloride (MC). Gwathmey and Clement (2010) evaluated effects of PPD and MC on plant growth and development, stem starch reserves and lint yield. Higher PPD in narrower rows tended to reduce boll number more than leaf area/plant, reducing the number of bolls/unit leaf area. However, boll distribution was more concentrated, implying more synchronous demand for photosynthate in 25 cm rows than in wider rows. Stem starch concentrations during boll filling were similar to, or slightly lower, in 25 cm rows than in wider rows. Findings are consistent with the hypothesis that availability of photosynthate may limit boll load at higher PPD, despite more leaf area/boll. Application of MC tended to reduce leaf area/ plant more than boll load, and to increase the number of bolls/unit leaf area. The MC treatment also concentrated the boll set on lower sympodia, increasing the synchrony of boll maturation and demand for photosynthate. Application of MC increased boll set percentage and decreased stem starch reserves slightly. Lint yields were increased by MC application. Findings support the hypothesis that MC benefits boll set and yield formation in narrow-row systems by reducing LAI. Source sink alteration with PPD and MC may be useful in future research to improve carbohydrate use and yield formation in cotton.

In China Yang *et al.,* (2014) recorded the highest cotton yield at 3.0 plants/m^{-2} due to more bolls/unit ground area (79 bolls/m^{-2}), while the lowest yield was obtained at 1.2 plants/m^{-2}. The results suggest that 3.0 plants m^{-2} would be the optimal plant density because it provides a better canopy micro environment.

Zhi *et al.,* (2014) analyzed the spatial distribution of photosynthetically active radiation (PAR) in a heterogeneous cotton canopy based on a geo-statistical sampling method in six planting density. They observed: 1) transmission within the canopy decreased with increasing density and significantly decreased from the top to the bottom of the canopy, but the greatest decreases were observed in the middle layers of the canopy on the vertical axis and closing to the rows along the horizontal axis; 2) the transmitted PAR (TPAR) of 6 different cotton populations decreased slowly and then increased slightly as the leaves matured, 3) the Leaf area index (LAI) was highly significant exponentially correlated with the intercepted PAR (IPAR) within the canopy; 4) and a highly significant linear correlation was observed between the accumulated IPAR and the biomass.

Considering the need to break yield stagnation in cotton, a new trial was formulated under All India Coordinated Research Project on Cotton from 2012-2013 onwards to evaluate compact genotypes at closer spacing in all the cotton growing zones of India. Preliminary trial results indicated encouraging results (Manickam *et al.*, 2014b) and the yield barrier in cotton can be broken by adopting compact varieties in all the cotton growing tracts of India, especially under rainfed situation.

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Inheritance of resistance against cotton leaf curl virus disease in *Gossypium hirsutum L.*

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Cotton is the most important cash crop of North India. *G. hirsutum* and *G. arboreum* species are commercially cultivated in this zone. During the year 2013, 2014 and 2015 Upland cotton suffers losses even up to 100 per cent in some areas mainly due to high incidence of cotton leaf curl virus disease (CLCuD) and white fly. This disease is caused by *Gemini* virus which is of considerable concern and is transmitted by whitefly. Cotton leaf curl virus disease is initially characterized by small vein thickening symptoms on young upper leaves of plants. Use of chemicals in controlling the whitefly is costly and not so effective. Moreover, it may be hazardous to men and environment. Therefore, development of a resistant variety /hybrid to this disease is the most effective, long term, less expensive and safe method. First step in this direction is screening and identification of resistant sources and their incorporation in the agronomical superior genotypes/varieties. For his purpose the study of inheritance of character in question is very essential. Inheritance of resistant genes for disease is still under discussion whether it is nuclear or extra-nuclear. Azhar *et al.*, (2010) reported that the resistance dependent on major genes (dominant genes) may lose quickly because of evolution of pathogen for these genes. An alternative approach is needed for partial resistance that depends on the recombination of minor genes. In the present screening and identification of resistant parents from germplasm and breeding material were identified. Crosses were attempted between resistant and susceptible parents to CLCuD. Six generations were developed for study of CLCuD inheritance.

In North India it is the most important cash crop of *kharif* season where only *G. hirsutum* and *G. arboreum* species are grown. About 95 per cent area of cotton is under *G. hirsutum*. Since last two decades cotton crop is very adversely affected by increasing incidence of cotton leaf curl virus disease. It was reported first in Nigeria (1912) on *Gossypium peruvianum* and *Gossypium vitifolia*, Sudan (1924), Tanzania (1926), Philippine (1959), Pakistan (1967) in Multan district on scattered *hirsutum* plants (Hussain and Ali, 1975). It was not well thought-out as a serious disease up to 1987 in Pakistan but appeared in epidemic form in 1992-1993 and it resulted in huge financial losses with the estimated value of $5 billion (US) from 1992-1997 (Briddon and Markham., 2001). It is very complicated disease and is very difficult to calculate the precise estimates of losses caused by it because the occurrence of CLCuD varies from year to year and also varies from area to area under cotton cultivation. The disease caused by geminivirus and the climatic condition of North India remained favorable during the year 2013, 2014 and 2015 for the growth and spread of whitefly. Cotton leaf curl virus disease is a serious threat to cotton production in this region. In India, cotton leaf curl virus disease was first reported in American cotton (*G. hirsutum*) in

Sriganganagar area of Rajasthan state during 1993 (Ajmera, 1994) and during 1994 it also appeared in Haryana and Punjab (Rishi and Chauhan,1994; Singh *et al.*,1994) states in *hirsutum* cotton.

The disease has appeared in an epidemic form during 1997 in the Rajasthan affecting an area of 0.1 million hectares (Anonymous,1998). Cotton leaf curl virus disease is initially characterized by small vein thickening type symptoms on young upper leaves of plants. These irregular thickenings gradually extend and coalesce to form continuous reticulation of small veins. The disease is further characterized by upward or downward curling of leaves and affected leaves become thick, leathery, brittle and greener than healthy leaves. Later formation of cup shaped or leaf laminar outgrowth called "enation" appear on the underside of the leaf. Some time more than one enation may appear. Use of chemicals in controlling the whitefly (the vector of this virus) is costly and it can't be controlled completely as single whitefly may infect to number of plants. Moreover, it may be hazardous to men and environment. Extensive uses of pesticides have also caused damage to soil quality and fertility (Dinham, 1993). Therefore, development of a resistant American cotton variety to this disease is the most effective, long term, less expensive and safe method to fight against this disease and to enhance the productivity of cotton. Research efforts to develop resistant varieties/hybrids through conventional/ biotechnological approaches along with cultural and management practices are in progress for effectively controlling this disease. Nature has provided cotton with traits like okra leaf type, gossypol glands and trichomes which confer non-preference to the insect pest infestation. Knowledge of genetic architecture of the trait in question to be improved is very important.

Disease incidence and economic losses : It is difficult to estimate the actual losses as it varies from year to year and cultivar to cultivar. Sometimes, cotton field have been found to show as much as 100 per cent damage and in recent years a recovery from disease incidence have been observed in later stages of maturity. The reduction in yield due to CLCuD incidence depends upon the time of infection and severity of disease. Monga *et al.*, 2004 reported that there was reduction in monopodia, sympodia, leaf size, inter node length and plant height as the disease grade increases. Yield losses due to this disease is reported to be 38 -60 per cent from Haryana, Punjab and Rajasthan. (Malathi *et al.*, 2004). Up to 20 per cent losses have been reported due to this disease in Sudan in *G.barbadense* var. 'Sahel'. Singh (2006) studied that on an average, there was 50.4 per cent reduction in number of bolls due to CLCuD infection, reduction of 42.9 per cent in the boll weight. Due to CLCuD the fibre length was reduced by 5.2 per cent, strength by 5.4 per cent, elongation by 10.0 per cent, uniformity by 2.2 per cent and micronaire value by 4.1 per cent in the diseased plants over healthy plants. Therefore, on the basis of four years studies they concluded that CLCuD infection adversely affects both seed cotton yield and quality. Similarly Khan *et al.,* (1995) and Akhtar *et al.*, (2009) measured losses inflicted by CLCuD by comparing yield and components (number of bolls / plant and yield / plant).

Selection of parents : Four parents including two resistant (H 1098-i and H 1117) and two susceptible (B 59-1678 and HS 6) to cotton leaf curl disease were chosen as the experimental material for the present study.

These four parents were used to develop four crosses, H 1098-i x B 59-1678 (R x S), H 1117 x HS 6 (R x S), H 1098-i x H1117(R x R) and B 59-1678 x HS 6 (Sx S). These crosses were designated as cross I, cross II, cross III and cross IV, respectively.

Development of basic generations : During *kharif*, 2011, the parents were identified to fulfil the objectives and F1 crosses between these parents, namely H1098-I, H 1117, B 59-1678 and HS 6 were made. The F1 hybrids and parents were raised during *kharif,* 2012. Each F1 was selfed to obtain F2 generation and simultaneously backcrossed to both of its parents to produce backcross generations (BC1and BC2). Fresh crosses were also made to obtain the F1 seed and all the parents were selfed to get their seeds for the next year. Thus, the experimental material finally comprised of six generations, namely P1, P2, F1, F2, BC1 (backcross to first parent P1)and BC2 (backcross to second parent P2).

Layout and design of the experiment : The experimental material comprised of six generations i.e. parents (P1 and P2), F1, F2 and back crosses (B1 and B2) of four crosses was grown in a compact family block design with three replications during *kharif*, 2013 at Cotton Research Area of CCS Haryana Agricultural University, Hisar. There was a single row of non segregating generations (P1, P2 and F1), 20 rows of F2, and 8 rows of each BC1 and BC2 generations. The length of each row was 6 m with spacing of 67.5 x 30 cm. In order to build up heavy inoculum pressure one row of highly susceptible line (HS 6) was planted at the periphery of the experimental area. Normal cultural practices were followed except no insecticidal spray was given for control of white fly (*Bemisia tabaci* Genn) population in the field. White fly is the vector of cotton leaf curl virus disease. Reaction of cotton leaf curl virus disease was recorded on all the plants in all the three replications.

Observation on cotton leaf curl virus disease (CLCuD) : Observation on CLCuD was recorded under field condition in each replication on all the plants of each of the non –segregating generations (P1, P2 and F1), backcross generations and the F2 generation. Disease was scored on 0 -5 grade depending upon the response to the cotton leaf curl virus disease CLCuD. Disease symptoms were recorded every fortnight and final observation was taken on September 15, 2013.

$$\text{Disease incidence (\%)} = \frac{\text{Number of diseased plants}}{\text{Total number of plants}} \times 100$$

0 = Immune, Complete absence of symptoms 0 per cent disease incidence

1 = Highly resistant, Very minute thickening of veins, 0-10 per cent disease incidence

2 = Resistant, Thickening of small group of veins, 10-20 per cent disease incidence

3 = Susceptible, Severe vein thickening and leaf curling developed at the top of the plant, 20-40 per cent disease incidence

4= Moderately susceptible, Severe vein thickening and leaf curling developed on the half of the plant canopy, 40-70 per cent disease incidence

5 = Highly susceptible, Severe vein thickening, leaf curling and full stunting of the plant, 70-100 per cent disease incidence.

Inheritance of resistance to cotton leaf curl disease : Due to very high incidence of CLCuD, complete absence of symptoms i.e. 0 % disease incidence was not observed in both the

resistant parents. Although they falls in resistant category. In both the resistant x susceptible crosses (H 1098 – i x B 59 – 1678 and H 1117 x HS 6) all the F_1 plants were resistant on the basis of per cent disease index for CLCuD indicated the complete dominance of resistance over susceptibility. F2 generations segregated nearly 15 resistant and 1 susceptible indicated two genes involvement and their segregation was also near to 3 resistant and 1 susceptible with role of some modifier gene was also not ruled out. Main reason for this confusing obsverations were unavailabilty of immune parents for such study. No complementary gene action was observed in susceptible x suscetible cross as all he plants of this cross were susceptible.

Genetical basis of cotton leaf curl virus disease: In the present study, resistance to CLCuD was dominant over susceptibility. Similar results have been reported by Azhar *et al., (*2010), Bachchan Kumar (2002) and Ali (1997). In F2 generation of these crosses, segregation for resistance gave a good fit to the phenotypic ratio of 15 (resistant): 1 (susceptible), which suggested that presence of either or both dominant genes produces resistance reaction. Backcross generation of resistance x susceptible cross with susceptible parent also segregated in 3 (resistant): 1(susceptible) ratio which further confirms that two recessive genes in homozygous condition were responsible for expression of disease and presence of dominant gene(s) at either locus or at both loci result in resistance. Resistant and susceptible parents used in the present study were diverse for two genes. Expression of resistance in parents H1117 and H 1098 – i having resistance to CLCuD was governed by two dominant genes in homozygous condition, whereas susceptibility to this disease in susceptible parents was governed by two recessive genes in homozygous condition. Ahuja *et al.,* (2007) made crosses among 30 germplasm lines of upland cotton. In 22 crosses, 4 types of segregation patterns were obtained in the F2 generations. A good fit for 15 (resistant):1 (susceptible), 13 (resistant):3 (susceptible), 9 (resistant):7 (susceptible) ratios indicated digenic control of the trait with duplicate dominant, dominant inhibitory, and duplicate recessive epistasis, respectively. Three gene controls with triplicate dominant epistasis was obtained in one of the crosses. Contrary to this, in the earlier reports, Saddig (1968) and Rehman *et al.,* (2005) found that resistance to CLCuD was controlled by a single dominant gene. This might be because of different genetic stock used in their study. To determine whether the genes for resistance to CLCuD in H 1117 and H 1098 – i were same or different, the cross between resistant (H 1117) x resistant (H 1098 – i) was attempted in F2 generation, no segregation was observed indicating that genes for resistance to CLCuD in H1117 and H 1098 – i were same. This was further confirmed by the backcross generations.The possibilities of obtaining resistant reaction by complementary interaction of genes responsible for susceptibility was explored by studying the cross between susceptible x susceptible (B 59 – 1678 x HS 6). All the plants in segregating as well as in non segregating generations of this cross were susceptible. This indicated that there was no complementary interaction between the genes for susceptibility in the susceptible parents used. Whereas the nuclear inheritance is under discussion, the extra chromosomal inheritance is also a secret and considered to be absent by Rehman *et al.,* (2005) but the presence of reciprocal differences in the cross LRA 5166 is advocated by Khan *et al.*, (2007).

Till now nothing can be concluded about

CLCuD behaviour, its appearance, correlation with environmental conditions, its losses etc. During *kharif*, 2015 in a highly susceptible variety (HS 6) to this disease infection appeared in early stage of growth i.e. in first fortnight of J with per disease index grade of more than 3 but this variety recovered tremendously by first week of October will produce a yield between 15 to 20 q/ha but it is not true for different years / location / sowing time of crop or some other factors amy be invloved.

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Development of high yielding better fibre quality varieties/hybrids of *desi* (*G. arboreum*) and American cotton (*G. hirsutum*) through conventional breeding for Marathwada region of Maharashtra

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ABSTRACT : Understanding the need of farmers for *rainfed* cotton cultivation, Vasantrao Naik Marathwada Krishi Vidyapeeth thoroughly involved in developing high yielding coupled with better *fibre* quality cotton genotypes varieties/ hybrids through conventional cotton breeding programme. As concerned to the desi cotton varieties, PA 32 (Eknath) , PA 141 (Namdeo), PA 183 (Sawata), PA 255 (Parbhani Turab) and PA 402 (Vinayak), PA 08 amd PA 528 which involved greater shift of deshi cotton varieties towards higher improvement in seed cotton yield whether by hirsutised cotton or by introgression of *hirsutum* traits into *arboreum* cultivars. . American cotton straight varieties PH 93 (Nagnath) , NH 452 (Renuka), NH 545 , PH 348 (Yamuna) ,NH 615 (Anusaya) and NH 635 have improved seed cotton yield and shift in fibre length from medium to long staple length. *Intra hirsutum* hybrids NHH 44, NHH 1, NHH 302, PHH 316 and NHH 250 have also shown their existence in hybrid cotton development. NHH 44 also the one of the milestone, from Cotton Research Station, Nanded had fulfilled the farmers requirement in *rainfed* as well as in *irrigated* areas not only upto Maharashtra but also in inter state and international level also. Two inter specific hybrids NHB 12 , *G. hirsutum x G. barbadence* origin and Pha 46, *G. herbacium x G. arboreum* were also developed by CRS, Nanded and CRS, Mahboob Baugh Farm, Parbhani , respectively. The continuous work to improve seed cotton yield and fibre properties in desi, American Cotton varieties and hybrids through conventional breeding method uplifts resource poor farmers rainfed cultivation under limited resources.

Cotton plays an important role in Indian farming and industrial economy of the county. Success of any crop breeding programme is based on the knowledge and availability of genetic variability for efficient selection.. As regards the development of American cotton, release of biotic and abiotic resistance, tolerance varieties and hybrids depend upon conventional breeding programme and gene pool availability. The importance of *des*i cotton has increased further as it is immune to cotton leaf curl virus (CLCuV) which has become the major disease of American cotton in the whole cotton north zone belt. *Desi* cotton can be successfully grown in CLCuV prone area where the incidence of this disease is relatively more. Though *Bt era* have made cumulative changes in cotton breeding progamme, we have to still not escape from conventional breeding programme.

In this context a summary on *desi* cotton *(G. arboretum*) varieties, American cotton (*G . hirsutum)* varieties and *intra hirsutum* hybrids released by cotton group of Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani is taken (Table 1).

[I] *Desi* cotton varieties : :

PA 183 (Sawata) : It is a derivative of three way cross of NA 39 x (C-C-1-1-3 x Lohit) followed by pedigree method of breeding at Cotton Research Station , Mahboob Baugh Farm,

Table 1 : List of the notified cotton varieties/hybrids developed by Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani.

Sr. No.	Name	Species	Notification	Year	Institution	Average yield (q/ha)	GOT (%)	Fibre length (mm)	Bundle strength (g/tex)	Micronaire value	Spinning potential (Counts)
1	2	3	4	5	6	8	9	10	11	12	13
I	***Deshi* varieties- *G. arboreum***										
1	PA 32 (Eknath)	*G. arboreum*	615-E	1980	CRS, MB farm, Parbhani	9-10	40	24-25	20.5	4.8	30^{s}
2	NA 48 (Rohini)	*G. arboreum*	295 (E)	9.4.1985	CRS, Nanded	12-14	38-39	25-26	20.5	4.8	30^{s}
3	NH 239 (Purnima)	*G. arboreum*	540 (E)	24.7.1985	CRS, Nanded	11-12	39	24-25	19.5	4.2	30^{s}
4	PA 141(Namdeo)	*G.arboreum*	615 (E)	17.8.1993	CRS, MB farm, Parbhani	11	39-40	26	21	5	30^{s}
5	PA 183(Sawata)	*G.arboreum*	1 (E)	1.1.1996	CRS, MB farm, Parbhani	12-13	38.3	26-27	20.52	4.56	30^{s}
6	PA 255 (Parbhani Turab)	*G.arboreum*	161(E)	04.2.2004	CRS, MB farm, Parbhani	14-15	38.1	27-28	22	4.5	40^{s}
7	PA-402(Vinayak)	*G.arboreum*	S.O.122 (E)	02.2.2005	CRS, MB farm, Parbhani	16	38.4	26-27	21.5	4.6	40^{s}
8	PA 08	*G.arboreum*	—	—	CRS, MB farm, Parbhani	16-18	37.0	27.70	21.3	4.9	30^{s}
9	PA 528	*G.arboreum*	—	—	CRS, MB farm, Parbhani	16-18	39.0	27.00	20.7	4.8	30^{s}
II	**American cotton varieties - *G. hirsutum***										
1	PH 93(Nagnath)	*G. hirsutum*	615 (E)	17.8.1993	CRS, Parbhani	10-12	38	24	19.5	4.8	20^{s}
2	NH 452(Renuka)	*G. hirsutum*	1 (E)	1.1.1996	CRS, Nanded	11-12	40-41	24-25	20.5	4	30^{s}
3	NH 545	*G. hirsutum*	161 (E)	04.2.2004	CRS, Nanded	12-15	39	24-25	19.2	4	30^{s}
4	PH 348(Yamuna)	*G. hirsutum*	S.O. 122(E)	02.2.2005	CRS, Parbhani	13-15	39	26	20.1	5.0	30^{s}
5	NH 615 (Anusaya)	*G. hirsutum*	S.O. 449(E)	11.2.2009	CRS, Nanded	12-15	39	28	20.6	4.1	30^{s}
6	NH 635	*G. hirsutum*	—	—	CRS, Nanded	12-14	35.5	29.25	19.65	4.15	45^{s}
III	**American cotton hybrids - *Intra hirsutum* hybrid (*G. hirsutum x G. hirsutum*)**										
1	NHH 44	*G. hirsutum xG. hirsutum*	295 (E)	9.4.1985	CRS, Nanded	10-12 (R) 22-23 (I)	3535	24-25 25-26	2021	4.54.0	30^{s}40^{s}
2	NHH 1 (Godavari)	*G. hirsutum x G. hirsutum*	540 (E)	24.7.1985	CRS, Nanded	12-15(R)) 22-23 (I)	34	27 –28	20.5	4.2	60^{s}
3	NHH 302	*G. hirsutum x G. hirsutum*	615 (E)	17.8.1993	CRS, Nanded	14-15	38	26-27	21	4.0	40^{s}
4	PHH 316(Ganga)	*G. hirsutum x G. hirsutum*	821(E)	13.9.2000	CRS, Parbhani	17	38	27-28	21.5	4.2	40^{s}
5	NHH 250	*G. hirsutum x G. hirsutum*	—	—	CRS, Nanded	14-16	35.4	27.7	20.8	4.1	50^{s}
IV	**(*G. hirsutum x G. barbadence*) hybrid**										
1	NHB 12	*G. hirsutum xG. barbadence*	639 (E)	17.8.1990	*CRS, Nanded*	10-15	33	30-32	22	3.5	80^{s}
V	**(*G. herbacium* x *G. arboreum*) hybrid**										
1	Pha 46	*G. herbacium x G. arboreum*	1 (E)	1.1.1996	CRS, MB farm, Parbhani	16	34	28-29	22	5	40^{s}

Table 2. Performance of PA 183 (Sawata) in comparison with PA 32 (Eknath) and PA 141 (Namdeo)

Sr.No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)			Per cent increase over	
				PA 183	PA 32	PA 141	PA 32	PA 141
1.	1989-1990to1993-1994	University breeding trials	14	1790	1464	1554	22.2	15.1

Table 3. Fibre properties of PA 183 (Sawata) in comparison with PA 32 (Eknath) and PA 141 (Namdeo)

Sr. No.	Year	Fibre parameters	PA 183	PA 32	PA 141
1.	1989-1990, 1991-1992 and 1992-1993	2.5 per cent Span length	28.7	25.2	26.2
		Uniformity ratio	49.6	48.3	50.6
		Micronaire	4.13	5.13	4.00
		Fibre strength (PSI)	9.1	9.0	8.96

Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani. The dwarf plants were selected in F_3 population of C-C-1-1-3 x Lohit and were crossed with NA 39 during 1984-1985. Selections were made for plants having dwarfness, earliness, longer staple length and higher ginning outturn. F_3 plant to row progenies were grown during 1985-1986 and selections were made among dwarf lines. Similaraly F_4 families were grown during 1986 -1987, F_5 during 1987-1988 and F_6 during 1988-1989. The selected lines no 183 (in F_7 generation) was tested in trials from the year 1989-90 (Anonymous,1995). PA 141 (Namdeo) and Eknath (PA 32) are similar to PA 183 for most of the morphological characters. On an average of 14 breeding trials conducted during 1989-1994, the variety PA 183 (1790 kg/ha) recorded 22.2 per cent and 15.1 per cent increase seed cotton yield over PA 32 (1464 kg/ha) and PA 141 (1554 kg/ha), respectively (Table 2).As per fibre properties this variety is *at par* with PA 32 and PA 141 (Table 3).

PA 255 (Parbhani Turab) : To develop productive long staple, early maturing and drought tolerant *desi* cotton variety suitable for rainfed region, the parent CJ 73 (Sanjay) having high bolling potential and drought tolerance was crossed at Cotton Research Station , Mahboob Baugh Farm, Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani with parent A 4 having big boll size . The F_1 was further back crossed with CJ 73. The selected plants of back cross progeny of (CJ 73 x A 4) x CJ 73 in F_3 generation was further crossed with parent NA 39 having excellent fibre qualities (28-29 mm mean fibre length) and early maturity (Anonymous, 2002 (a)). The variety PA 255 (1324 kg/ha) recorded 21.10 per cent enhanced seed cotton yield than PA 183 (1093 kg/ha). In AICCIP trials , on an average of 11 trials conducted PA 255 (1219 kg/ha) recorded 16.10 per cent higher seed cotton over AKA 8401 (1050 kg/ha) (Table 4). Staple length of PA 255 (28.3 mm) had found 1.7 mm longer than PA 183 (26.6 mm) (Table 5). The strain PA 255 (28.7 mm) had more than 2.0 mm longer 2.5 per cent span length (fibre length) than the check PA 183 (20.3 mm). Other technological paroperties were *at par* with the check (Table 6).

PA 402 (Vinayak) : To raise the productivity and to improve fibre qualities of desi cotton to bring them to an level of *hirsutum* cotton

Table 4. Performance of PA 255 in comparison with PA 183

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)			Per cent increase over
				PA 255	PA 183	AKA 8401	PA 183
1.	1991-1992 to 2001-2002	University breeding trials	51	1324	1093	—	21.10
2.	1993-1994 to 1997-1998	AICCIP trials Central and South Zone	11	1219	—	1050	16.1

Table 5. Ginning outturn (%) and staple length (mm) of PA 255 in comparison with PA 183

Sr. No.	Year	Trial	No. of trials	Ginning outturn (%)			Staple length (mm)
				PA 255	PA 183	PA 255	PA 183
1.	1994-1995 to 1999-2000	Cotton arboreum Breed 02 (a), Proj. Br. 22 and 24	14	38.2	38.3	28.3	26.6

(*hirsutization* of *desi* cotton) as well as to introgress big boll size and fibre traits of *Gossypium hirsutum* (American cotton) into *G. arboreum* (*desi* cotton) (Introgression through interspecific hybridization) Polyploidy was induced at Cotton Research Station , Mahboob Baugh Farm, Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani in *G. arboreum* variety PA 85 /85. This is the first *introgressed* variety in the *G. arboretum.* Owing to limitations due to ploidy level in such *interspecific* hybridization, polyploidy was induced in *Gossypium arboreum* (2n=26) variety PA 85/85 by colchicines treatment. The resultant autotetraploid (2n= 52) was crossed with *hirsutum* variety NH 239 (Pornima) having 2n=52 chromosomes. Resultant interspecific F_1 having 27 percent pollen fertility was backcrossed with original induced autotetraploid PA 85/85. From segregating population of backcross progeny, the variety PA 402 have been isolated by adopting pedigree method (Anonymous,2003). Probably , this is the first variety developed through introgression in *desi* cotton. The variety, PA 402 had high productivity under rainfed condition. On an average of 34 breeding trials conducted for six years (1997-1998 to 2002-2003), the variety PA 402 (1010 kg/ha) recorded higher yield potential with 22.7 percent increase over popular variety PA 255 (823 kg/ ha). The variety PA 402 also had higher yield

Table 6. Fibre technology data of PA 255 in comparison with PA 183

Year	Trial	No. of trials	2.5 per cent span length (mm)		Uniformity ratio (%)		Micronaire value		Fibre strength	
			PA 255	PA 183	PA 255	PA 183	PA 255	PA 183	PA 255	PA 183
1994-1995 to 1998-1999	Cotton arboreum Breed 02 (a), Proj. Br. 22, 24	04	28.7	26.3	48	49	4.7	4.7	20.3	20.5

Table 7. Performance of PA 402 in comparison with PA 183 and PA 255

Sr. No.	Total trials	Trial name	Year	Mean seed cotton yield (kg/ha)				Per cent increase over		
				PA 402	PA 183	PA 255	NH452 /Rajat	PA 183	PA 255	NH452 /Rajat
1.	34	Station , University level and SMVT trials	1997-1998 to 2002-2003	1010	805	823	—	25.4	22.7	—
2.	11	NATP and TMC	2001-2002 to 2002-2003	1411	983	1167	721	43.5	20.9	95.7

Table 8. Ginning outturn and staple length of PA 402 in comparison with PA 183 and PA 255

Sr. No.	Total trials	Trial name	Year	Ginning outturn (%)			Staple length (mm)		
				PA 402	PA 183	PA 255	PA 402	PA 183	PA 255
1.	16	University level, SMVT, NATP and TMC	1997-1998 to 2002-2003	38.4	37.7	37.6	26.1	26.3	26.9

Table 9. Fibre properties of PA 402 in comparison with PA 183, PA 255 and *hirsutum* varieties

Sr. No.	Year	Fibre parameters	PA 402	PA 183	PA 255	NH 452/ PKV 081/ LRK 516
1.	2001-2002 and 2002-2003	2.5 per cent Span length	25.2	24.7	26.6	24.3
		Uniformity ratio	50.0	49.5	47.6	48.6
		Micronaire value	4.7	4.5	4.5	4.2
		Fibre strength g/tex	21.2	21.7	21.7	18.3

than *hirsutum* variety NH 452/ Rajat. On an average of 11 trials conducted for two year (2001-2002 to 2002-2003), the variety PA 402 (1411 kg/ ha) gave near 100 per cent enhanced seed cotton yield over *hirsutum* variety NH 452/ Rajat (Table 7). In addition , the variety PA 402 possesses fibre qualities at par with quality of *arboreum* PA 183 and PA 255 and superior than *hirsutum* variety NH 452 (Table 8 and 9).

PA 08 : *G. arboreum,* a naked seeded variety released during joint Agresco, having tolerant to drought, sucking pest, bacterial blightand grey mildew.

PA 528 : *G. arboretum,* a superior ginner variety, released during Joint Agresco, 2015 , having tolerant to sucking pests, bacterial blight and grew mildew.

[II]American cotton varieties :

PH 93 : American cotton variety developed by Cotton Research Scheme, Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani known as Nagnath, developed by hybridization followed by pedigree selection in cross J 207 x AC 738 (Anonymous,1990). On an

average of 40 trials conducted during 1985-86 to 1988-1989, PH 93 (1628 kg/ha) recorded 27.6 per cent higher seed cotton over Purnima (1275 kg/ha) (Table 10).

NH 452: NH 452 a stable variety of American cotton was released by Cotton Research Station, Nanded under Vasantrao Naik Marathwada Krishi Vidyapeeth , Parbhani . Due to early maturity and tolerance to biotic and *abiotic* stress, this variety performed well in areas with medium soils and scanty rainfall. *G. hirsutum* variety NH 452 was found superior than cotton varieties LRA 5166 and PH 93 (Anonymous,1994). On the basis of average of 23 varietal trials conducted during 1990-91 to 1993-1994 at multi location , NH 452 (1808 kg/ha) recorded 18.9 % and 17.6 % higher seed cotton yield over checks PH 93 (1525 kg/ha) and LRA 5166 (1471 kg/ha). In 14 AICCIP trials conducted during 1991-1992 to 1993-1994, the variety NH 452 (1256 kg/ha) recorded 21.2-23.1 per cent increased seed cotton yield over local check PH 93 (1036 kg/ha) and LRA 5166 (1020 kg/ha) respectively (Table 11).

Table 10. Performance of PH 93 in comparison with Purnima

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)		Per cent increase over Purnima
				PH 93	Purnima	
1.	1985-1986 to 1988-1989	University ,Multi location, Co ordinated varietal trials	40	1628	1275	27.6

Table 11. Performance of NH 452 in comparison with PH 93 and LRA 5166

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)			Per cent increase over	
				NH 452	PH 93/LC	LRA 5166	PH 93/LC	LRA 5166
1.	1990-1991to1993-1994	University breeding trials	23	1808	1525	1471	18.9	17.6
2.	1991-1992to1993-1994	AICCIP trials central zone	14	1256	1036	1020	21.2	23.1

NH 545 : NH 545 has compact sympodial plant type and is found suitable for agronomic manipulation (high plant density). This genotype developed by Cotton Research Station, Nanded under Vasantrao Naik Marathwada Krishi Vidyapeeth , Parbhani ,found free from *parawilt* and gray mildew under field conditions and is resistance to cotton leaf curl virus (Anonymous, 2002 (b)). Due to superior yield performance over NH 452 , this variety is recommended for cultivation in Marathwada region. On an average of 33 trials conducted over 7 years (1994-95 to 2000-2001) , NH 545 recorded 22.5 % higher seed cotton yield over check , NH 452 and on an average of 15 trials conducted over 04 years (1994-1995,1995-1996,1996-1997 and 2000-2001) recorded 12.9 higher seed cotton yield over check hybrid NHH 44. Also recorded 19.6 % increased seed cotton yield over PHH 316 (646 kg/ha) in 17 trials conducted under university level during 1997-1998 to 2000-2001 (Table 12). NH 545 has stapal length of 24.7 mm , strength 22 g/tex and micronaire of 3.5 (Table 13)

PH 348 : American cotton variety PH 348

Table 12. Performance of NH 545 as compare to NH 452, NHH 44 and PHH 316

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)				Per cent increase over		
				NH 545	NH 452	NHH 44	PHH 316	NH 452	NHH 44	PHH 316
1.	1994-1995to2000-2001	University breeding trials	33	967	789	—	—	22.5	—	—
2.	1994-1995to2000-2001	AVT., MKV 04 b, NATP	15	1040	—	898	—	—	12.9	—
3.	1997-1998to2000-2001	AVT., MKV 04 b, NATP	17	773	—	—	646	—	—	19.6

Table 13. Fibre properties of NH 545 as comparison to NH 452

Sr, No.	Variety	Fibre parameter			
		2.5 per cent SL (mm)	UR	Micro	Strength (g/tex)
1	NH 545	24.7	48	3.5	22.00
2	NH 452	24.3	49	3.9	22.00

released as long staple variety having high productivity, excellent fibre qualities and tolerance to drought for cultivation under *rainfed* eco-system by Cotton Research Scheme, Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani (Anonymous,2002 (c)). On an average of 21 trials conducted over 5 years, PH 348 recorded 33.88 % higher seed cotton yield over check NH 452 and 14.4 % higher seed cotton yield over NH 545 in 12 trials. In AICCIP trials, total 06 trials, PH 348 (894 kg/ha) recorded 38.8 % and 103.6 % increased seed cotton yield over local check NH 452 (644 kg/ha) and Zonal Check LRA 5166 (439 kg/ha) (Table 14). The variety PH 348 had 2 mm longer staple length than NH 545.

Table 14. Performance of PH 348 in Marathwada.

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)			Per cent increase over	
				PH 348	NH 452	NH 545	NH 452	NH 545
1.	1997-1998 to 2001-2002	University breeding trials	21 12	744 880	556 -	- 769	33.88 -	- 14.4
2.	2001-2002	AICCIP trials	06	894	644 (LC)	439 (ZC-LRA 5166)	38.8 (LC)	103.6 (ZC-LRA 5166)

Similarly PH 348 had more ginning percentage than NH 545 (Table 15).

NH 615 : Sustained efforts to develop superior varieties of American cotton were made to develop NH 615 at Cotton Research Station, Nanded. This variety showed constituently superior performance for yield, fibre quality and ginning out turn over previous existing improved cultivars of American cotton. Besides yield advantage, NH 615 is found tolerant to drought, major sucking pests and important diseases and well adopted to dryland area of Central Zone which includes states of Maharashtra, Madhya Pradesh, Gujrat and part of Odisha (Anonymous, 2009). The variety NH 615 also proven its strength of cultivation under high density planting and also proven novel in organic farming

Table 15. Ginning out turn (%) & staple length (mm) of PH 348 in comparison with NH 545.

Sr. No.	Year	Trial	Mean GOT (%)		Mean staple length (mm)	
			PH 348	NH 545	PH 348	NH 545
1	1999–2000 to 2001-2002	University trial, SMVT and AICCIP Trials	38.4	38.0	28.0	26.0

Table 16. Performance of NH 615 in Marathwada, Maharashtra and Central Zone.

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)			Per cent increase over	
				NH 615	NH 545	PH 348	NH 545	PH 348
1.	2002-2003 to 2001-2002	University breeding trials, SMVT and TMC Trials	31	1233	1012	1069	21.8	17.0
2.	2003-2004 to 2001-2002	AICCIP trials central zone(Maharashtra Trials)	26	1388	1202 (LC)	-	16.0 (LC)	-
3.	2003-2004 to 2001-2002	AICCIP trials central zone	24	1358	1039	862 (ZC-LRA5166)	30.7 (LC)	57.5 (ZC-LRA5166)

specially in Vidharbha region. This variety was developed by pedigree method of selection from segregating material of cross between NH 545 x JLH 1492. On an average of 31 trials conducted across Marathwada Region over 5 years, NH 615 (1233 kg/ha) recorded 21.8 and 17 per cent higher seed cotton yield over checks, NH 545 (1012 kg/ha) and PH 348 (1069 kg/ha), respectively. On the basis of 26 trials conducted across Maharashtra State, NH 615 recorded 16 per cent higher seed cotton and lint yield respectively over local check. On the basis of 24 trials conducted across different locations of Central Zone over four seasons, NH 615 recorded 30.7 and 57.5 % higher seed cotton yield over local and Zonal check, (LRA 5166), respectively (Table 16).

NH 635 : American cotton varieity, NH 635, was released and recommended by Joint Agresco, 2015 for cultivation in Maharashtra State under rainfed conditions. This variety was also derivative developed by pedigree method of selection from segregating material of cross between NH 545 x JLH 1492 (Anonymous,2015).. On an average of 81 trials conducted during 2005-2006 to 2014-2015 throughout Maharashtra State , the variety NH 635 shown 12.76, 21.38, 26.14, 41.05 and 24.38 per cent higher seed cotton yield over checks PH 348 ,NH 615, PKV Rajat, AKH 8828 and Phule 688 respectively (Table 17). The proposed variety is tolerant to bacterial blight and *Alternaria* leaf spot. The variety is also found tolerant to sucking pests .As for quality parameters, the variety has recorded mean fibre length of 29.25 mm ,fibre strength of 19.65 g/tex and Micronaire of 4.15. The mean seed oil content of NH 635 is 17.30 per cent and biomass of 2.9 t/ha .The proposed variety NH 635 is also found suitable for planting under high density planting system (HDPS) . Hence the test variety, NH 635 is recommended for release in Maharashtra State.

[III] *Intra hirsutum* cotton hybrids :

NHH 44 : *Intra hirsutum* hybrid NHH 44 is a cross between BN 1 x AC 738 and released

Table 17. Summary of NH 635 tested in station/SMVT/AICCIP trials conducted during 2005-06 to 2012-13

Sr. No.	Trial Name	Year	No. of Location	Seed cotton yield (kg/ha)							
				Variety				Checks			
				NH 635	PH 348	NH 615	PKV Rajat	AKH 8828	Phule 688	Local checks (AICCIP trials)	Zonal check (NH 615) (AICCIP trials)
1	Station/ University trials	2005-2006 to 2014-2015	30	1130	1027	945	—	—	—	—	—
2	SMVT trials	2012-2013 to 2014-2015	25	1084	—	963	918	821	931	—	—
3	AICCIP trials	2009-2010 to 2012-2013	26	1259	—	—	—	—	—	1072	1082
		Total/ Mean	**81**	**1158**	**1027**	**954**	**918**	**821**	**931**	**1072**	**1082**
		per cent increase over check			**12.76**	**21.38**	**26.14**	**41.05**	**24.38**	**8.02**	**7.02**

Table 18. Performance of NHH 44 as compaire to H 4 .

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)		Per cent increase over
				NHH 44	H 4	
1.	1977-1978to1982-1983	University varietal trials	23	2317	1807	28.3
2.	1980-1981to1982-1983	Agronomy trials	13	2416	1836	31.5
3.	1970-1981to1982-1983	T.S.F./T.C.D. trials	11	1559	1214	28.4
4.	1980-1981to1981-1982	Cultivators field trials	54	1242	947	27.1

by Cotton Research Station, Nanded (Anonymous,1984).. A novel hybrid having rejuanation quality of flowering , drought tolerant , favored by farmers under ranifed and as well as irrigated farming. On an average of 23 university level varietal trials conducted during 1977-1978 to 1982-1983, the hybrid NHH 44 (2317 kg/ha) recorded 28.3 per cent increased seed cotton yield over check H4 (1807 kg/ha). In 13 Agronomy trials conducted during 1980-1981 to 1982-1983, NHH 44 (2416 kg/ha) recorded 31.5 per cent increased seed cotton yield over check H4 (1836 kg/ha) under irrigated condition. During 1980-1981 to 1982-1983, in T.S.F./ T.C.D. trials (total 11 trials), the NHH 44 (1559 kg/ha) recorded 28.4 per cent increased seed cotton yield over H4 (1214 kg/ha). About 54 cultivators field trials conducted during 1980-1981 to 1981-1982 , NHH 44 (1242 kg/ha) recorded 27.1 per cent increased seed cotton yield over H4 (947 kg/ha) (Table 18).

PHH 316 (Ganga) : I*ntra hirsutum* hybrid PHH 316 , a cross combination between female PH 93 and male PH 325 (selection from PKV 081) , developed at Cotton Research Scheme, Parbhani (Anonymous,1997). On an average of 17 trials conducted at university level, the hybrid PHH 316 (1490 kg/ha) recorded 20.5 per cent higher seed cotton yield over check NHH 44 (1236

Table 19. Performance of PHH 316 in Marathwada, and Central Zone.

Sr. No.	Year	Trial	No. of trials	Seed cotton yield (kg/ha)			Per cent increase over	
				PHH 316	NHH 44	NHH 302	NHH 44	NHH 302
1.	1991-1992	University	17	1490	1236	—	20.5	—
	to1996-1997	breeding trials,	15	1489	—	1164	—	27.9
2.	1993-1994	AICCIP trials central zone	10	1716	1407 (CC) (H 6)	1524 (LC)	14.0 (CC) (H 6)	

kg/ha) and in 15 university level trials, NHH 44 (1489 kg/ha) recorded 27.9 per cent increased seed cotton yield over check NHH 302 (1164 kg/ha) (Table 19). Due to its 20 per cent higher yield potential , 3 to 3.5 per cent higher ginning out turn and 3 to 4 mm longer staple length over NHH 44 (Table 20) and free from parawilt, it satisfy respectively to farmers , traders and textile personnel's.

Table 20. Fibre properties of PHH 316 as comparison to NHH 44

Sr, No.	Variety	Fibre parameter			
		2.5 per cent SL (mm)	UR	Micro	Strength (g/tex)
1	PHH 316	27.8	49	3.5	19.8
2	NHH 44	23.9	48	3.6	17.9

NHH 250 : *Intra hirsutum* hybrid NHH 250, developed by Cotton Research Station, Nanded , recommended and released during Joint Agresco, 2015 (Anonymous,2015 (b)).. On an average of 34 trials conducted during 2006-07 to 2014-15 throughout Maharashtra State , the proposed hybrid NHH 250 shown 23.77, 59.22, 71.60, 47.40 and 29.06 per cent higher seed cotton yield over checks Bunny, NHH 44, PKV HY 4, PKV HY 5 and Phule 492 respectively (Table 21). The proposed hybrid is tolerant to Bacterial blight and *Alternaria* leaf spot. The hybrid is also found tolerant to sucking pests. As for quality parametes, the test hybrid has recorded mean

Table 21. Summary of yearwise performance of *Intra hirsutum* (H x H) test hyrid NHH

Sr. No.	Trial Name	Year	No. of Location	Seed cotton yield (kg/ha)								
				Variety	Checks							
				NH 250	Bunny	NH 44	NHH 206	PKV HY4	PKV HY5	Phule 492	Local checks (AICCIP trials)	Zonal check (NH 615) (AICCIP trials)
1	Station/ University trials	2006-2007 to 2009-2010	7	1509	1341	862	—	—	—		—	—
2	SMHT trials	2008-2009 to2014-2015	10	1215	906	884	1043	810	943	1077	—	—
3	AICCIP trials	2010-2011 to20012-2013	17	1445	—	-	—	—	—		1275	1166
		Total Mean	**34**	**1390**	**1123**	**873**	**1043**	**810**	**943**	**1077**	**1275**	**1166**
	Per cent increase over check			**23.77**	**59.22**	**33.26**	**71.60**	**47.40**	**29.06**	**9.01**	**19.21**	

250 during 2006-2007 to 2012-2013 under rainfed condition.

fibre length of 27.7 mm ,fibre strength of 20.8 g/ tex and Micronaire of 4.10. The mean seed oil content of NHH 250 is 17.87 per cent and biomass of 2.57 t/ha. Hence the test hybrid, NHH 250 is recommended for release in Maharashtra State.

Release of such high yielding varieties/ hybrids of G. arboreum and *G. hirsutum* at state/ national level helped to increase average productivity of the region as well as State. The most popular *intra hirsutum* hybrid NHH 44 dominated cotton cultivation scenario of the country for two decades and slowly moved out of chain with the introduction of *Bt* cotton. Similarly, most of the parents of varieties of *G. arboreum* and *G. hirsutum* released by Vasantrao Naik Marathwada Krishi Vidyapeeth, Parbhani are used as a donor for desease/pest resistance in breeding programme at various centres. Similarly, they are used as parent in producing hybrid combinations available in the market by private sector. The variety NH 615 of *G. hirsutum* also found suitable for HDPS (High Density Planting System) particularly under shallow soils. The glory of these non bt varieties/hybrids may come back in coming years as the conversion programme of the straight varieties/hybrids are already initiated under Public Private Partnership with Maharashtra State Seed Corporation Limited, Akola. The *Bt* versions of *G. hirsutum* varieties/hybrids may helped to fulfill needs of the cultivators of the region. The *G. arboreum* varieties PA 528, PA 08, PA 402, PA 255 are performing very well in the region and area may be expecting to increase in coming years especially organic cotton cultivation.

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Functional finishing of cotton textiles

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Cotton is one of the wonderful fibres gifted to human kind especially in the tropical countries of the world. Cotton textiles are comfortable to wear, possess good absorbency, heat conductivity, tensile strength, abrasion resistance, absence of static problems etc. It is a versatile fibre that can be molded to suit various implied functions. Cotton and other natural cellulosic fibres are chemically reactive unlike most synthetic fibres which are not very reactive and some are considered inert. The reactive groups on the cotton molecule permit permanent attachment of these functional compounds. The surface of the fibre is polar and hydrophilic, which makes the fabric comfortable during wear and useful for absorbent applications such as towels. Furthermore, the fibre has a large surface area and is porous somewhat like a sponge. Other properties of this fibre, which make these modifications possible, are the optimum degree of crystallinity and a useful range of fibre micronaire (denier) and fiber length.

With the advent of science and technology, a new area has been developed in the realm of textile finishing either improving the process or helping to achieve new functional properties which are not possible with conventional finishing. The nanotechnology, plasma enhanced chemical vapour deposition (PECVD) and Layer by Layer (LbL) assembly, use of Phase Change Materials (PCM) are some of the new techniques that have resulted in revolutionary changes in the area of textile finishing. Thus, it is possible today to alter cotton fabric and make it thermal regulatory, self-cleaning, antimicrobial, UV protective and so on.

The nanotechnology which deals with the materials at nano stage has tremendous applications in textile field especially in finishing. Application of nano finishing chemicals provide effective ultra-thin finished surface that enable the textile to exhibit functional property without altering its hand and feel. Nano finishes such as stain resistance, antimicrobial, controlled hydrophilicity / hydrophobicity, antistatic, UV protective, wrinkle resistant and shrink proof abilities can be exploited using this technique for a range of technical textile applications such protective clothing, medical textiles, sportswear, automotive textiles etc.

They are generally emulsified into either nanomicelles, made into nanosols or wrapped in nanocapsules that can adhere to textile substrates easily and more uniformly. Since 275 nanoparticles have a large surface area to volume ratio and high surface energy, they have better affinity for fabrics. Therefore these finishes are more durable, effective and do not adversely affect the original handle and breathability of the fabric.

Plasma polymerization enables deposition of very thin nanostructured coatings (< 100nm) via gas phase activation and plasma substrate interactions. This dry and ecofriendly technology offers an alternative to replace wet chemical process for surface modification

(finishing) of textiles. Plasma polymerization can impart a wide range of functionalities such as water repellency, hydrophilicity, dyeability, conductivity and biocompatibility due to the nano scaled modification of textiles and fibers.

The principle underlying the use of phase change materials is applicable to textiles for providing thermal comfort in garments. When a rise in temperature occurs, the PCM microcapsules react byabsorbing heat and storing this energy in the liquefied phase change materials. When the temperature falls again, the microcapsules release this stored heat energy and the phase change materials solidify again. It is now possible to impregnate cotton material with microcapsules containing a small amount of PCM that helps in improving the thermal insulation of fabric through absorption of energy during heating and release of it during cooling. PCM microcapsules could be directly incorporated into fibres and foams or typically applied to fabrics as a coating.

Layer by layer assembly (L-b-L) is a unique technique for the fabrication of composite films and deposition of coatings with nanometer precision. Nano coating of cotton substrate using L-b-L process enables imparting various functional properties on cotton textiles such as antimicrobial , self cleaning, hydrophilicity / hydrophobicity etc. Cotton fibers offer unique challenges to the deposition of nanolayers because of a unique cross-section as well as chemical and physical heterogeneity of its surfaces. Cationic cotton surface has been successfully coated with alternate layers of anionic and cationic polyelectrolytes, *i.e.* poly (sodium 4-styrene sulphonate) and poly (allylamine hydrochloride) using L-B-L technique (Joshi.M).

This paper mainly focusses on nano finishes on cotton textiles that provide soil repellent and antimicrobial properties required for healthcare textiles.

The most important aspect of health care is to provide a conducive and comfortable environment to facilitate the quick recovery of the patient. Microbial contamination of surfaces, including textile fabrics, can lead to information on infections. These hospital acquired infections prolong the healing of patients, and cause potential risks for serious illnesses. The growing public health awareness of the pathogenic effects, malodours and stain formations caused by microorganisms, has increased the need for antibacterial materials in many health care applications. Based on this need, an attempt was made to develop a fabric suitable for health care applications with nano finishes to impart antimicrobial and stain resistant properties

Hundred per cent cotton twill weave fabric and polyester cotton blend was selected for the white coats. For bed sheets, hundred per cent cotton 20s sheeting fabric was selected. The nano form of zinc pyrothine derivative as antimicrobial chemical and nano fluorocarbon derivative as stain resistant chemical were selected in 6 different combinations.

In experimental research design was followed for this study. In the Phase I, the antimicrobial nano chemicals and Stain repellent nano chemicals were optimised and the best performing concentration were taken forward to develop a dual nano finish for the Phase II. In the Phase III the wear study was conducted to understand the human handling factor on performance of the finish.

Assessment of antifungal and antimicrobial activity of dual finished nano fabric : The assessment of antifungal activity of the nano finished fabric against *Aspergillus niger* was evaluated using AATCC – 30 – 2004.

qualitative and quantitative assessment of antibacterial activity of the nano finished fabric was evaluated as per AATCC standard – 147 – 2004 andAATCC 100- 2004 respectively against *Escherichia coli* and *Staphylococcus aureus*. Analysis for Stain resistance assessment of the nano finished fabric was carried out by AATCC 130- 2000 toward blood, medicine and oil stains. The laundering procedure was followed as per AATCC 135 – 2004.

The hospital uniforms and sheets were made from the finished fabric and subjected to washing after each usage in a hospital. The fabric functional properties were assessed after 5 washes, 10 washes and 15 washes.

The antifungal and antimicrobial activity of the samples treated and untreated (control) are presented in table 1.

The fungus *Aspergillus niger* was not developed on all treated fabrics irrespective of the fibre content. The antifungal activity of the treated fabrics against this fungus was excellent even after 15 washes. It indicates that the nano finish applied on fabric is ideal and suitable for adoption in the field of healthcare textiles.

The zone of inhibition against *E.coli* was recorded as 5 mm in case of cotton coat fabric and sheeting fabric. Around 4 mm ZOI was observed for polyester cotton blend fabric. For S. aureus, 6 mm ZOI was recorded for cotton coat fabric and sheeting fabric and 5.5 mm for polyester cotton blend fabric. It was observed that with increase in number of washes the zone of inhibition decreased slightly. The sheeting fabric maintained its antimicrobial activity against *E.coli* even after 15 washes. Further it was noted that the antimicrobial activity was higher against S. aureus compared to *E. coli* for all the three fabrics.

Assessment of stain resistance for blood stains, oil stains and medicine stains : Assessment of stain resistance for the dual nano finished fabric after wear study was carried out according AATCC -130- 2000. The samples were rated on a scale from grade 5 to 1 by comparison between the residual stain on the test specimen with the stains on the stain release replica. Grade 5 represented the best stain removal and Grade 1 to the poorest stain removal.

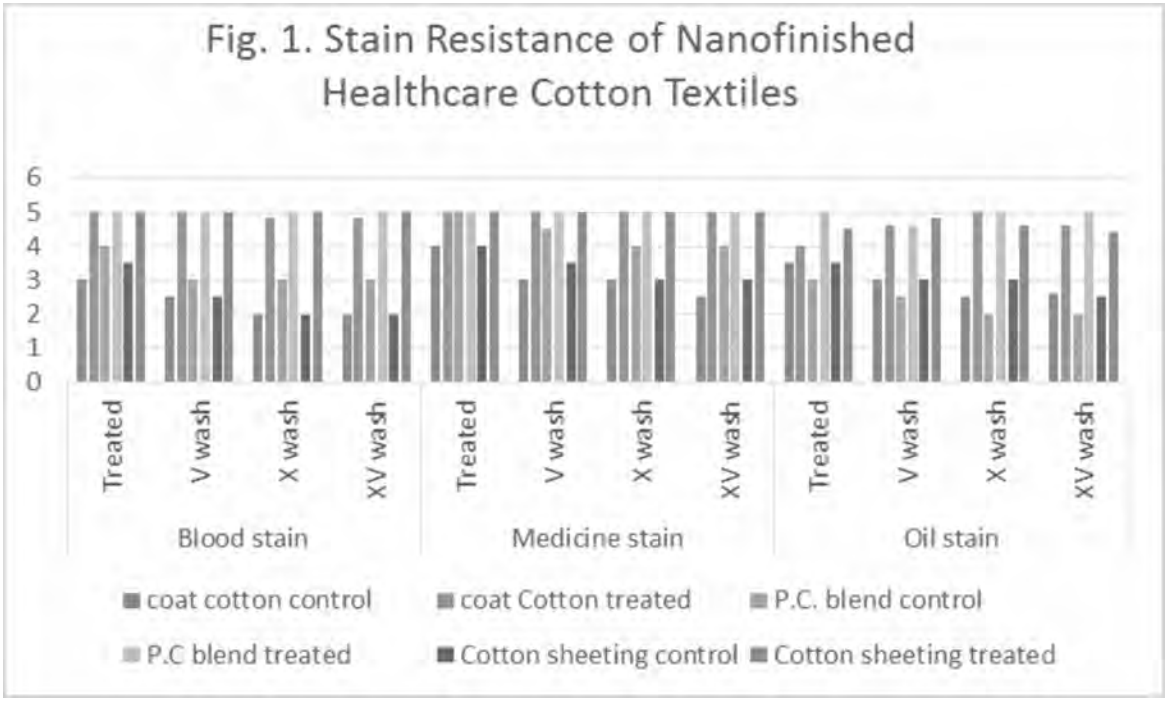
Table 2 depicts the stain resistant property of the dual nano finished fabric after the wear study.

It was observed that the three selected

Table 1. Antifungal and antimicrobial activity of dual finished nano fabric after the wear study against *Aspergillus niger E. coli* and *S. aureus* (zone of inhibition)

Samples	Antifungal activity *Aspergillus niger*				Antimicrobial activity (ZOI)** *E.coli*				*S.aureus*			
	Treated/ un-treated	V wash	X wash	XV wash	Treated	V wash	X wash	XV wash	Treated/ un-treated	V wash	X wash	XV wash
Cotton control	4*	4	4	4	0	0	0	0	0	0	0	0
Cotton treated	0	0	0	0	5	4	4.5	4	6	5	4.5	4
P.C blend control	4*	4	4	4	0	0	0	0	0	0	0	0
P.C blend treated	0	0	0	0	4	3	2	2	5.5	4	3.5	2.5
Cotton sheeting control	4*	4	4	4	0	0	0	0	0	0	0	0
Sheeting treated	0	0	0	0	5	5	5	5	6	5.5	5	4.5

*untreated controls **Zone of Inhibition



fabrics had excellent stain resistance towards blood stains. It was also observed that the fabrics maintained excellent rating (grades) after fifteen washes of wear study as clearly represented in figure 1. Sheeting fabrics in emergency and maternity wards require resistance to blood stains which is a crucial factor for their further use. This showed that the nano finish formulation selected was an appropriate choice for the health care worker's uniform and sheeting.

Similar observations were made with medicine stains. The fabrics maintained excellent rating even after fifteen washes of wear study as clearly indicated in Fig. 2. This showed that the nano finish formulation selected was a pertinent choice for the health care worker's uniform and fabrics.

The resistance towards oil stain was observed to be excellent. The stains were graded as 4 in case of cotton coat fabric, grade 4.5 in sheeting and grade 5 in polyester cotton blended fabric. Not much difference was found in the rating of all the three fabrics after the wear study

Table 2. Stain resistant property of dual finished nano fabric for the wear study towards blood, medicine and oil stains

Samples	Blood stain (grade)				Medicine stain (grade)				Oil stain (grade)			
	Treated/	V wash	X wash	XV wash	Treated	V wash	X wash	XV wash	Treated	V wash	X wash	XV wash
Cotton control	3	2.5	2	2	4	3	3	2.5	3.5	3	2.5	2.5
Cotton treated	5	5	4.8	4.8	5	5	5	5	4	4.6	5	4.6
P.C blend control	4	3	3	3	5	4.5	4	4	3	2.5	2	2
P.C blend treated	5	5	5	5	5	5	5	5	5	4.6	5	5
Cotton sheeting control	3.5	2.5	2	2	4	3.5	3	3	3.5	3	3	2.5
Sheeting treated	5	5	5	5	5	5	5	5	4.5	4.8	4.6	4.4

and after subjecting to fifteen washes.

The cost of production of these experimental dual nano finished fabrics was estimated to be 20 to 25 per cent higher than the unfinished fabrics. But it can be reduced to 10 to 12 per cent in bulk production.

CONCLUSION

Cotton textiles after treatment with nano dual finish containing nano chemicals exhibited good antimicrobial and stain resistance. As the finish is durable upto 15 washes, the finished fabric will be an ideal healthcare textile that control the infections in hospitals.

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Development of GIS and GPS based spatial cotton fibre quality maps

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ABSTRACT : A case study was carried out to design and develop Geographical Information System (GIS) and Geographical Positioning System (GPS) based spatial cotton fibre quality maps for Nagpur district of Vidharbha region of Maharashtra. Spatial database and spatial fibre quality maps for parameters such as 2.5 percent span length, uniformity ratio, fineness, strength, elongation, ginning percentage, short fibre content, degree of reflectance and degree of yellowness were developed. Spatial distribution, classification and characterization of the district based on the fibre quality were carried out to provide site specific information. The quality features of the cotton grown in Nagpur district was analyzed by visualizing the spatial maps. The 2.5 per cent span length of the cotton was observed to fall in two classes, long and extralong, and the uniformity ratio into three classes, average, good and excellent. The fineness of the cotton could be categorized into three classes, very fine, fine and average. The strength of the cotton was found between good and very good. It was observed that cotton grown in moderately deep to deep clayey soil are of extralong staple with average fineness, good strength and lower short fibre content. Cottons grown in very shallow to shallow loamy soils were of long staple, and fine with average strength and higher short fibre content. Cotton produced in plains was found to have higher staple, fineness and strength compared to those produced at higher elevations. It is concluded that spatial maps would be highly useful to traders, ginners, researchers and industry for efficient planning and decision making process.

Keywords: Cotton, fibre quality, GIS, GPS, spatial maps

With the global shift towards market economics, the need for timely, reliable and location specific information has become more important. A comprehensive knowledge of the spatial cotton quality parameters is of fundamental importance to cotton traders and industry for efficient planning of their business.Cotton is being traded in the market based on its variety and grade. First hand spatial information related to cotton grown and its quality parameters is not available. Even if available, it is represented in tabular forms and it is difficult to interpret and visualize the variations and assess the availability of particular quality of cotton in the district, region or state.

Recently emerged technologies like Geographic Information System (GIS) and Geographical Positioning System (GPS) are widely used as spatial analysis tool for effective and efficient means of data acquisition, data storage and retrieval, manipulation and analysis and output generation (Baily, 1990). A GIS offers many advantages for spatial data management and presentation, including a structured representation for data, data management functions, and most importantly, visualization of data. Spatial simulations can take advantage of large quantities of data previously digitized as GIS layers. (Mccauley, 1999). It provides continuous surfaces from point data. Spatial distribution, classification and characterization

of the region are possible with GIS based on any phenomenon. It helps to analyze trends over time, and spatially evaluate impacts caused by development. GIS allows developing decision making processes much faster (Bantalan *et al.,* 2000)

A GIS combines geographic mapping capabilities with a database management system. A GIS database consists of a set of data layers, usually referencing a common coordinate system, which describe different thematic or quantitative information. Applying GIS to the process of preparing crop estimates has improved accuracy while lowering costs (Fourie, 2008).

GIS based spatial analysis was conducted and the best locations for harvest collection centres were determined, based on the shortest and least cost path of delivery by the farmer. The maps produced have proven to be critical tools for the field officers for route planning when conducting field visits. This has led to a considerable cut in the cost of production (Felix, 2011)

To circumvent this problem, a case study was carried out and to develop GIS and GPS based spatial fibre quality maps for cotton grown in Nagpur. Spatial fibre quality maps would be useful for classification of the area under cotton based on fibre quality. Spatial maps provide site specific information. As visual interpretation of information in the form of maps allows finding variations quickly, these maps would be highly useful to traders, ginners, policy makers and researchers for systematic planning of their interest for the particular area or the region.

MATERIALS AND METHOD

Seed cotton samples from spatial locations were collected by using GPS from the cotton growing area of Nagpur district by following stratified sampling method. In stratified sampling method the population is divided into subpopulations (strata) and random samples are taken of each stratum in a number proportional to the stratum's size when compared to the population. The longitude, the latitude and the elevation of the locations were noted. A GPS instrument- Magellan Triton 200 was used for recoding the spatial locations. The study was carried out during the crop season 2009-2010.A total of 306 samples were collected from Nagpur district. The spatial locations were widely distributed and representative of the study area with predominantly grown cotton varieties. Secondlyspatial locationswere recorded from thesampling area by using GPS (Fig. 1).

The samples were ginned on the Lilliput gin developed by CIRCOT. The ginned samples were tested on High Volume Instrument (HVI 900) for measurement of fibre quality. Digitization and geo-referencing of the toposheets of Nagpur district were carried out. The noncotton growing area was delineated from the cotton growing area.

The spatial database and the spatial fibre quality maps for parameters namely 2.5 percent span length, uniformity ratio, fineness, strength, elongation, ginning percentage, short fibre index

Table 1. Talukawise samples collected from Nagpur district

Sr. No.	Taluka	Spatial locations
1	Parsioni	53
2	HIngna	36
3	Narkhed	49
4	Ramtek	19
5	Katol	41
6	Saoner	48
7	Kalmeshwar	43
8	Kamptee	17

Fig 1. Recording of spatial location by using GPS.

(SFI), degree of reflectance (Rd) and degree of yellowness (+b) were prepared. Spatial distribution, classification and characterization of the Nagpur district based on each fibre quality were carried out. These spatial fibre quality maps were correlated with the soil maps, such as soil depth and soil texture.

RESULTS AND DISCUSSION

GIS based spatial fibre quality database and maps : Fibre quality and GPS data of spatial locations obtained from the sampling area was analyzed and depicted in Table 2.

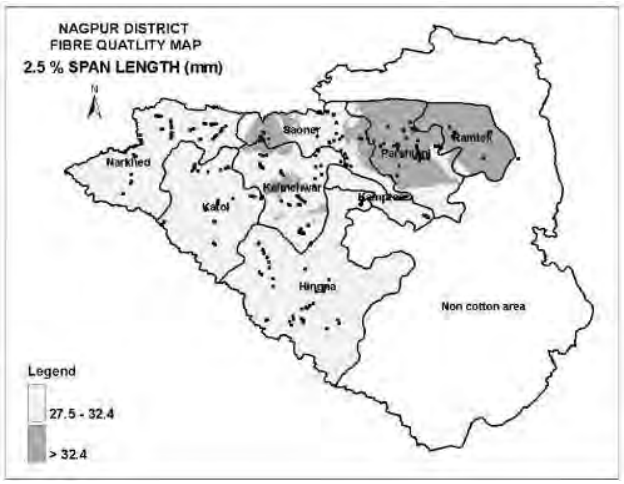
The spatial database and maps for fibre quality parameters was developed using ARCGIS software. Significant variation in fibre quality parameters among the different Talukas were observed in Nagpur district. Spatial database and spatial fibre quality maps were prepared. The spatial maps of Nagpur district for different fibre quality parameters are shown below (Fig. 2a to f).

Spatial distribution, classification and characterization of the Nagpur district based on the fibre quality were carried out. The quality features of the cotton grown in Nagpur district was analyzed by visualizing the spatial maps (Table 3).

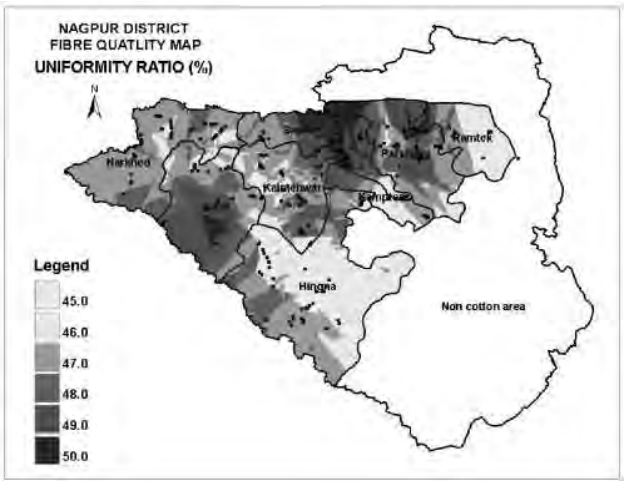
The features of the cotton grown in Nagpur district that was observed by looking at the maps were, staple of the cotton was found to be long and extralong, uniformity ratio was observed to be from average to excellent, fineness between very fine to average and the strength good to very good.

Correlation of spatial fibre quality maps with soil maps : The soil depth, the soil texture and the elevation maps of the Nagpur district were prepared in the GIS environment. The soil depth maps of the cotton growing area were delineated from the noncotton growing area. The soil in thestudy area was found to be clayey and loamy based on the depth of soil (Fig. 3). Further based on the texture, the soil in study area was found to be deep, moderately deep, moderately shallow, shallow, very shallow and extremely shallow (Fig. 4). The elevation of the study area was found between 245 to 470 m.

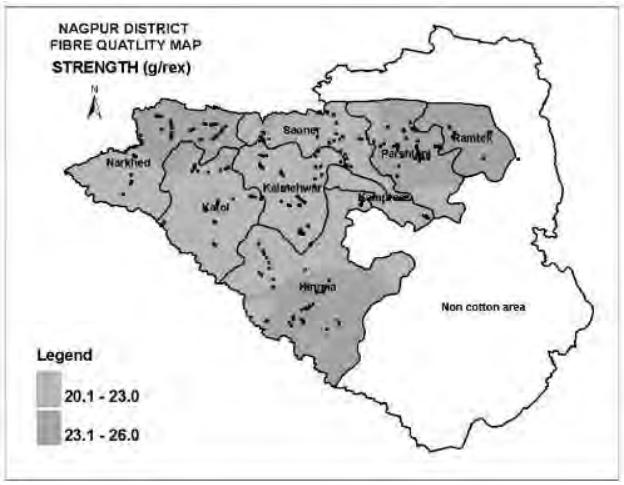
The cotton produced in Nagpur district was found to be of long and extra long staple. 2.5 per cent span length was found between 28.1 to 33.9 mm. Cotton produced in the deep clayey soil was found to be extralong type. In loamy soil, the long staple cotton was mostly grown. At higher elevations, fibre length of the cotton was found to lower compared to the cottons in the plains. The fineness of cottons produced in this region was found between 2.9 to 4.2 mic. In moderately deep to deep clayey soil, fineness was found to be of average category and ranged between 4.0-4.9 mic. In shallow loamy soils, the fineness was observed in the fine category and ranged between 3.0-3.9 mic. At higher elevations in clayey soil, the fineness of the cotton was found below 3.0 mic. At higher elevations fineness of the cotton was found to be comparatively lower than the



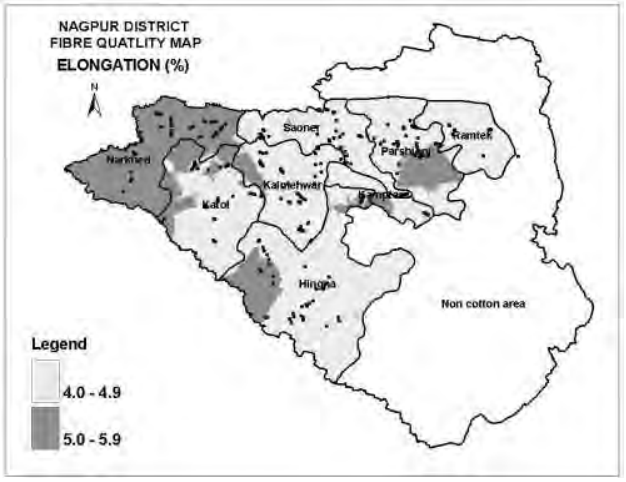
(a) Spatial map for 2.5 percentspan length



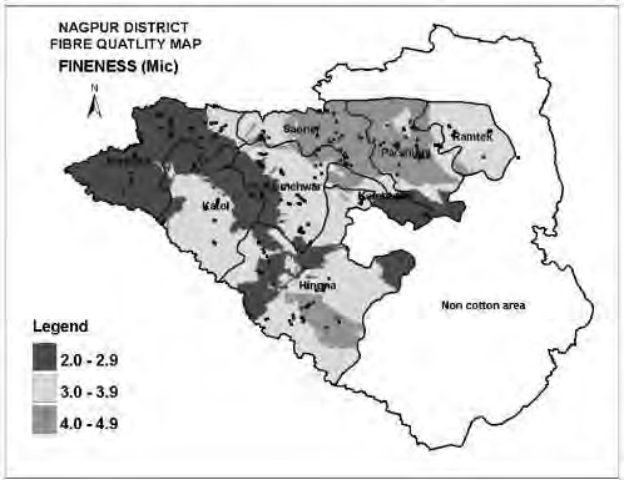
(d) Spatial map for uniformity ratio



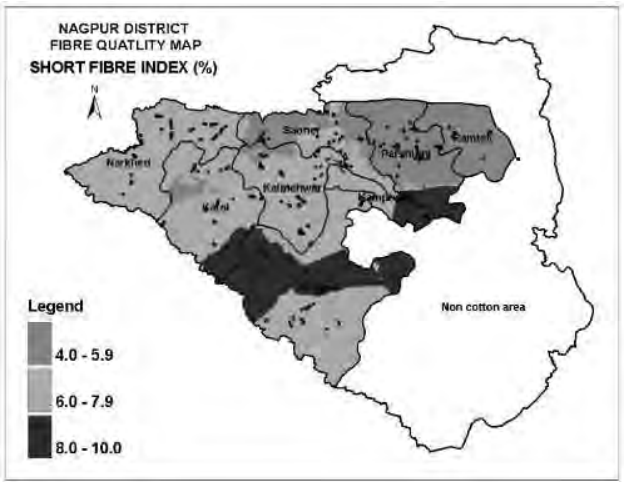
(b) Spatial map for fibre strength



(e) Spatial map for elongation



(c) Spatial map for fineness



(f) Spatial map for short fibre content

Fig 2. Spatial fibre quality maps of Nagpur district

Table 2. Variation in the fibre quality parameters in the Nagpur district.

Parameter	Min	Max	Average
Logitude	78.24.17 E	79.23.04 E	—
Lattitude	20.54.33 N	21.50.55 N	—
Location elevation(m)	204	324	318
GP (%)	27.2	40.9	33.1
Fibre length (mm)	24.0	36.4	30.7
Uniformity ratio (%)	39	54	48
Fineness (mic)	2.2	5.6	3.6
Strength (g/tex)	17.7	27.5	23.6
Elongation (%)	3.6	6.4	5.0
SFI (%)I	3.5	14.6	6.1
Rd (%)	54.3	84.8	77.4
+b (%)	4.6	10.8	7.7

Table 3. Fibre quality of cotton as visualized form spatial maps

Fibre quality parameter	Classification	Range
2.5 per cent span length (mm)	Extra long	> 32.5
	Long	27.5-32.5
Uniformity ratio (%)	Excellent	> 47
	Good	45-46
	Average	44-45
Fineness (mic)	Very Fine	< 3.0
	Fine	3.0-3.9
	Average	4.0-4.9
Strength (g/tex)	Very Good	> 26.1
	Good	23.1-26.0

cottons in the plains.

The fibre strength of the cottons produced in this region was found to range from 20.2 to 26.0 g/tex. In moderately deep to deep clayey soil, strength was found to be of good category ranging from 23.1 to 26.0 g/tex. In shallow loamy soils, the strength of the cotton was observed in the average category and ranged from 20.1 to 23.0 g/tex. At higher elevations, strength was found to be a bit lower than the cottons in the plains.

The per cent fibre elongation was found better in clayey soils than loamy soils. In clayey soils, it was found to range from 5.0 to 5.9 per cent and in loamy soils 4.0 to 4.9 per cent. The short fibre content of the cotton produced in this region was found to vary from 4 to 10 per cent. Cottons in loamy soils found to have short fibre content of 6.0 to 7.9 per cent and in clayey soils 4.0 to 5.9 per cent. Cottons produced in clayey soils were found to have lower short fibre content than the cottons in loamy soils.

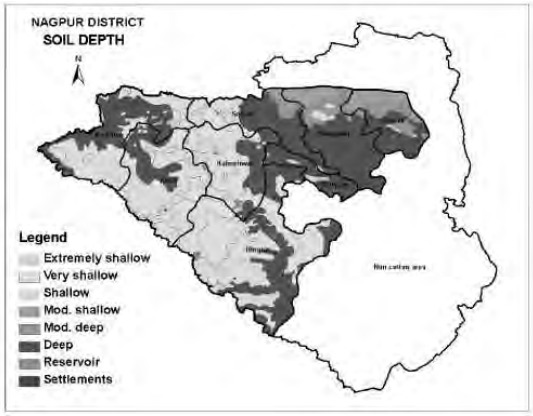


Fig 3. Soil depth map for Nagpur district

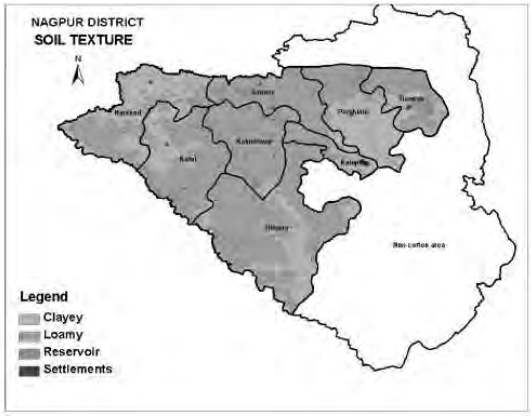


Fig 4. Soil texture map for Nagpur district

Table 4. Cotton fibre quality with respect to soil type

Soil type	Fibre Quality
Moderately deep to deep clayey	Extra-long staple, average fineness, good strength and lower short fibre content.
Very shallow to shallow loamy	Long staple, fineness of fine category, with average strength and comparatively higher short fibre content.

CONCLUSIONS

- The spatial distribution, classification and characterization of the Nagpur district based on the fibre qualitywerefound to be very effective in visualization and interpretation of the data.
- The 2.5 per cent span length of the cotton was observed to fall in two classes *i.e.* long and extralong, uniformity ratio into three classes *i.e.* average, good and excellent, fineness in three classes *i.e.* very fine, fine and average and the strength was found between good and very good.
- Definite trend in fibre quality with soil type was noted.
- Spatial maps found to be useful to traders, ginners, researchers and industry for efficient planning and decision making process as it provides site specific information.

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Need for conservation agriculture in cotton

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Cotton, a major commercial crop, is grown on a wide range of soil types and climatic conditions and occupies nearly 12 m. ha area. About 65% of the area is rain dependent. Rainfall ranges from 500-1200 mm in the rainfed regions. Thus, soil moisture is a major constraint. Soils supporting cotton in the rainfed areas are Vertisols (Vertic intergrades), Inceptisols and Alfisols. Both Alfisols and Vertisols have different potentials and problems but both need organic C improvement and soil moisture conservation measures to restore their productivity.

Why to increase soil organic matter and how can we achieve it? : Frequent tillage operations are resorted to keep the land clean. It is a common sight to see a clean field of cotton free of weeds as well as any organic debris. Manure is rarely applied due to short supply. Thus, the soils are depleted of the organic C reserves. Furthermore, intensive tillage operations done on the farm leads to impairment of soil physical properties. As a result, the productive capacity of the soil is lowered resulting in demand for greater external inputs. Most often we hear about the lowering of factor productivity and increased cost of cultivation. How can we get out of this rot? Conservation Agriculture (CA) is the option for overcoming the present limitations in crop production.

Conservation Agriculture (CA) : The Food and Agriculture Organization (FAO) defined CA as a resource saving agricultural crop production that strives to achieve acceptable profits together with high and sustained production levels while concurrently conserving the environment.

CA has several benefits and differs in the time-scale in which such benefits are visible. Short-term benefits or benefits that are realized immediately include improved water infiltration, reduced evaporation, reduced soil and water loss. Those benefits that are seen in the medium and long-term are improved soil organic C, improved biological activity, and improved crop yields. The three steps that form the basis of CA are as follows;

1. Adding organic matter and maintaining soil cover
2. Adjusting crop rotation and/or
3. Reducing tillage operations

The above steps in effect address the issue of improving soil organic C. CA is a strategy that focuses on resource conservation. It refers to a system of raising crops without disturbing the soil while retaining crop residues or soil surface. CA offers farmers an array of practices, but at its core are three interlinked principles that can be applied in a variety of combinations to meet the needs of resource poor famers. There are a number of challenges that need to be addressed to make CA practicable and adoptable; changing mindset of farmers, retaining crop residues and utilizing it on the farm due to its competing uses, weed control issues and nutrient management.

CA is now used to distinguish a more sustainable agriculture from the erstwhile

narrowly defined 'conservation tillage'. It removes the emphasis from the tillage component alone and addresses a more enhanced concept of the complete agricultural system. Conservation tillage a more widely-used terminology denotes soil management systems that result in at least 30 per cent of the soil surface being covered with crop residues after seeding of the subsequent crop. To achieve this level of ground cover, conservation tillage normally involves some degree of tillage reduction and the use of non inversion tillage methods. Experiences on the benefits of CA and other issues are discussed specifically with reference to cotton. Further, it is important to note that just as one size of apparel does not fit all, CA has to be tailored according to the cropping pattern, agro climatic zone and the soil type.

Adding soil organic matter and maintaining a cover : The best manner, in which organic matter can be maintained in soil, is by direct application of good quality manure. However, this resource is in short supply. Furthermore, cotton is considered a low residue crop that may not provide sufficient surface residue to reduce erosion and protect the soil. To overcome the problem of short supply of organic manure, maintaining a soil cover is the best option. Thus, crop residue management is a critical part of CA systems. This can be done by growing of cover crops or inter crops within the wide row spaced cotton common in the rainfed areas or recycling of the crop residue of the previous crop in the irrigated north zone.

Four options available for crop residue mulch in cotton based systems are (i) mulch of previous crop residue, (ii) growing biomass producing crops, (iii) intercropping with short duration legumes, (iv) importing mulch from surrounding areas.

Mulch of previous crop residue : In the north zone, where double cropping is a predominant system, wheat crop residue can be recycled in cotton and cotton crop residues in wheat. However, cotton stalks are of poor quality than leguminous residues, because of their high lignin content, low C/N ratio (Blaise and Bhaskar, 2003) and therefore, could cause problems of N immobilization. However, trials from Sirsa, Sriganganagar and Ludhiana indicate incorporation of cotton and wheat crop residues improved productivity of both the crops. Incorporation of shredded wheat straw in the soil on sandy loam soil at Sriganganagar in Rajasthan state with one disc + two cultivator produced higher seed cotton yield of cotton (Nehra *et al.,* 2005).

In rainfed areas, continuous monocropping of cotton is a common practice because the quantity of rainfall does not permit double cropping. After picking, about 1.5-2 Mg/ha of cotton crop residue is available in the form of stalks and leaves. These are considered waste materials and are disposed off by burning. Farmers need to be advised that even if a small amount of residue is retained on the farm, it would increase SOC.

Intercropping/cover crops: Organic carbon is replenished when crop residue is recycled, apart from additional yield obtained from an intercrop. If an inter-crop is a legume, benefits of nitrogen fixed can be accrued. Saving of nearly 20-100 kg N/ha were reported (Blaise, 2011; Rochester and Peoples, 2005). In Australia, Rochester and Peoples (2005), reported cotton following wheat required 83 kg N/ha and there

was no need to fertilize cotton with nitrogenous fertilizer when vetch was grown after wheat. Vetches added nearly 230 kg N/ha to the soil when incorporated as a green manure, thus reducing dependency on fertilizer-N.

Growing biomass producing crops : The experiment conducted at CICR, Coimbatore, indicated that that sowing of finger-millet in off season by using available moisture (or summer rainfall) as bulk crop and *in situ* incorporation (45 days period) recorded significantly higher seed cotton yield (1402 kg/ha) of RCH 2 *Bt* as compared to control (fallow-cotton) (1192 kg/ha). Nayak (2004) reported that biomulch of sunnhemp showed maximum yield in rainfed cotton. The positive effect of sunnhemp biomulching may be attributed to good weed smother in the early stage, in addition to green manuring effect after incorporation. Adequate soil moisture availability and nutrient uptake under mulched environment were responsible for enhanced growth and yield. Spreading loppings from *Sesbania* spp obtained from 2 m dense rows after 10 cotton rows, in the entire field.

Importing mulch : Alternatively, green loppings of L*eucaenea* or *Sesbania* or *Crotolaria* can be grown on bunds around cotton and the loppings can be cut and spread. These systems are very simple but are labour intensive and improvement of soil structure and nutrient recycling are limited.

In the rainfed regions, the basic purpose of retaining a soil cover or residue mulch is to retain some of the available precipitation that is otherwise wasted. Sarkar *et al.,* (2007) under rainfed systems showed that soil surfaces with organic mulch can form a barrier against evaporation and can alter the microclimate. Consequently, soil moisture depletion occurs at a slower rate under organic mulches than under bare soil conditions.

Adjusting crop rotations : Diversification through crop rotation is an important CA principle and cotton is intercropped with diverse crops in different zones. Crop diversification tactics such as companion planting, barrier cropping, cover cropping and trap cropping are suggested as sustainable methods for managing crop pests (Hooks and Johnson, 2003). Continuous mono-cropping has become a common feature and is blamed for the ecological imbalance (create problems in soil, hydrology and biotic environments). Diversification is readily possible in irrigated areas as well as assured rainfall areas. However, in rainfed regions, the cropping system options are limited. Cotton can be rotated or inter-cropped with N fixing legumes. Legumes for intercropping during the kharif season are green gram, black gram, soybean, groundnut, cowpea, clusterbean etc. Options for intercropping in the later phase of the crop growth coinciding with the winter season are chickpea, garden pea, beans, forage legumes such as alfalfa and clovers. The N fixed by these legumes can be utilized by cotton. The prevalent cotton based systems in India are given in Table 1.

Well planned and designed cropping systems can avoid problems such as increased soil compaction, perennial weeds, and break pest and disease cycle. For instance, nematodes can severely affect cotton. Although, nematicides are available, for an effective level of nematode control, nematicides are expensive, toxic to humans and animals. Rotating cotton with a cereal reduces the population of nematodes

Table 1. Dominant cotton based cropping systems

State	Cotton based cropping system
North India	
Punjab, Haryana, Rajasthan	Cotton - Wheat (I), Cotton - Mustard (I)*
Central India	
Gujarat, Maharashtra, MP	Monocropped Cotton (R), Cotton + Black gram or Green gram or groundnut or soybean (R), Cotton - Sorghum (2 year) (R)
South India	
Andhra Pradesh	Monocropped cotton (R/I), Cotton-Rice (I), Cotton-Chilli (2 year) (I), Cotton- Tobacco (2 year I)
Tamil Nadu	Monocrop Cotton (R/I), Rice-Cotton (I), Rice-Rice-Cotton(I), Cotton-Sorghum(I), Cotton-Pulse-Cotton(I)
Karnataka	Monocrop cotton (R/I), Cotton-Wheat(I), Cotton+Chilli/ groundnut/black gram/ green gram (R)

*R: rainfed; I: irrigated

(Hake *et al.,* 1991). However, if rotated with soybean that shares similar nematodes, the problem can get aggravated. Rotating cotton with non-host crops of pink boll worm can reduce the infestation and need for insecticides. Cotton when rotated with a cereal crop reduces the problem of the noxious broadleaf weeds in cotton that are difficult to control. Rotating crops with differing rooting patterns also provide the benefits of different soil profiles being explored for moisture and nutrients. All these benefits are well identified. Presently the need is to integrate the crops providing the most complementary benefits.

In north India, cotton-wheat and cotton-mustard are common double cropping systems followed. The cotton-wheat system occupies nearly 1.7 million hectares in north Indian states of Punjab, Haryana and Rajasthan (Mayee *et al.,* 2008). In these cropping systems, although huge amount of crop biomass is produced, the crop residues are removed and destroyed by burning. As a result, SOC loss and water repellence in soil layers have been observed (Singh *et al.,* 2005). Consequently, productivity of cotton-wheat is on a decline (Jalota *et al.,* 2008) in spite of the widespread cultivation of the *Bt* hybrids. Retaining crop residues of both cotton and wheat residues have been reported to improve the productivity of cotton wheat grown on the sandy soils of Rajasthan (Nehra *et al.,* 2005). Das *et al.,* (2014) reported a reduction in water requirement by 14 per cent and improvement in the system productivity by 48 per cent where crop residues were retained compared to those without. Adoption of a legume during the *rabi* season such as chickpea, lentil, peas is a good practice as it reduces the reliance on nitrogen fertilizer. A new concept that is coming up is 'relay cropping cotton and wheat'. By taking up sowing of wheat in the inter-row spaces of cotton, timely sowing of wheat is ensured. Consequently, wheat yields do not decline as the crop is planted timely. Further, there is an advantage of getting a full crop of cotton. Taking up an 'extended crop of cotton' is another possibility. In China, Zhang *et al.,* (2007) observed relay cropping of cotton in wheat rows had land use advantage that was greater than monocultures. Further, resource use efficiency was also better in the relay intercropping systems (Zhang *et al.,* 2008). Recent studies in

Pakistan, too, indicate a benefit of intercropping cotton and wheat (Sultan *et al.,* 2012).

In central India, the general practice has been to cultivate long-duration pigeon pea as well as sorghum. The latter is now not practiced due to its poor market price. Cotton + pigeonpea strip intercropping is a traditional cropping system in central India. This practice is followed in about 1.5 million hectares in central. Although several options of N fixing legumes are available, inter cropping is not widely practiced. A major reason for the non adoption of intercropping, in central India, is that it interferes with inter row cultivation and weed control is difficult. Therefore, soil disturbance in the form of inter-row cultivation (hoeing) is necessary to control weeds emerging later during the crop growth. CA integrates herbicide for weed control. Herbicides currently available on the market have made post-emergent weed management easy in cotton. The current herbicides that are available for weed management are given in Table 2.

Availability of such selective herbicides has the potential to facilitate adoption of intercropping acceptable to the farmers. Soybean has recently been introduced in a big way in assured rainfall areas of central zone. Furthermore, introduction of Bt cotton with shorter duration, has opened up the possibility of double cropping in the traditionally mono-cropped assured rainfall areas with chickpea, safflower or lentil on residual moisture. This technology has the potential to generate more residues. However, use of the right inoculants for the legume seed is an important aspect if the benefits of nitrogen fixed has to be accrued.

Hulugalle *et al.,* (2007) reported that compaction increased in dryland Vertisols with cotton-based crop rotations after conversion from conventional to permanent raised beds. This compaction was less under rotations which cotton was followed by wheat or chickpea crop.

Herbicide tolerant (HT) cotton became commercially available in 1997 in the USA. Presently three HT cottons available are RR Flex

Table 2. Herbicide for weed management

Name of the herbicides/group	Method of application	Dose (kg a.i./ha)
Diuron	Pre emergence	1.0
Pendimethalin	Pre plant incorporation	1-1.5
Quizalofop ethyl	Early post emergence 20-25 DAS	0.05
Fenoxaprop methyl	Early post emergence 20-25 DAS	0.1-0.125
Propaquizafop	Early post emergence 20-25 DAS	0.1-0.125
Pyrithiobac Na	Early post emergence 20-25 DAS	0.125 to 0.25

(Roundup Ready Flex), Glytol tolerant and Dowstrike. In the USA, 78 per cent of the US cotton farmers who adopted conservation tillage practice have done so specifically because of herbicide resistant cotton varieties .and it has since been readily accepted by growers across the globe. Its potential benefits include the opportunity to reduce or eliminate soil-applied herbicides and to reduce total herbicide use, more effective weed management in conservation tillage systems, greater rotational crop flexibility, capability to control previously uncontrollable weeds etc. The greatest benefit to growers is the broad spectrum weed control and the convenience of post-emergence (over the top) application to cotton without crop injury.

This technology is not yet available in our country. Use of such non-selective herbicides in HT cotton would reduce the possibility of diversified inter-cropping and strip cropping systems that are in vogue at present. In addition there is a potential of some weeds become resistant to the herbicide.

Reduced tillage : Presently, intensive tillage operations are done to control weeds as well as to prepare a firm seed bed. There is a concern that cotton crop needs aeration and thus, frequent inter-row tillage operations are done. Contrary to the conventional tillage practices followed; CA applicable tillage systems are (1) non inversion and minimum tillage, (2) ridge planting or ridge furrow system and (3) strip tillage or zonal tillage (tillage only in seeding zone).

In the north zone where cotton–wheat is practiced, seedbed is prepared by two disking followed by two cultivators and planking. Crop residues are not incorporated in the field. The sticks of cotton are pulled out, removed from the field and used as fuel. In the subsequent crop of wheat, similar tillage operations are repeated once again that has lead to substantial loss of carbon and nutrients in soil. As a result, productivity of the cotton–wheat system has become static or started declining and is showing signs of fatigue (Jalot *et al.,* 2008). Reduced tillage (*i.e.* one disking, one cultivator and one planking) in cotton and minimum tillage (one planking only) in wheat were found sufficient from the view point of soil disturbance to sustain yield and apparent crop water productivity in cotton wheat system. This package of tillage operations increases net monetary saving and has a wide scope for adoption in cotton–wheat system by the farmers of Punjab in India and other countries where intensive tillage is practiced. Additionally, bed and furrow planting of cotton is finding favour with farmers due to savings in irrigation water and related benefits of improved use-efficiency of applied fertilizers, reduced soil crusting, etc. Jalota *et al.,* (2008), in a 2-year study, observed that yields were 23–39% higher in tillage treatments than minimum-tillage. Similarly in cotton production in the Tennessee Valley, USA, Schwab *et al.,* (2002) in a 4 year study reported 10% lower yields on zero tillage than for in-row subsoiling. In the the Mississippi Delta, USA (Pettigrew and Jones, 2001), an 11 per cent lower cotton yield under zero tillage compared with conventional tillage practices was observed. Similar results were reported for the semi arid region of Pakistan on a sandy clay loam (Ishaq *et al.,* 2001). In north India, where cotton is entirely irrigated, it is the low organic C content that limits crop productivity. In such situations, the adoption period of this system may take longer compared with permanent beds or conventional tillage systems. As long term investigations show (5-7 years), these problems reduce with improvement of soil quality (Gupta *et al.,* 2006). More recently, Das *et al.,* (2014) reported low yields in the zero till systems in the first two years, but subsequently, crop productivity increased compared to the conventional tillage systems.

In central and south India where cotton is grown on the Vertisols, frequent tillage operations are done. Conventional tillage (CT) involves mould board ploughing along with four to six inter row cultivations. Inter row cultivation (*daura*) is done for weed control besides 2-3 hand weeding depending on the weed intensity. Excessive tillage operations performed provide an effective control of weeds but also accelerate oxidation of organic matter. With the integration of herbicides, the number of tillage operations can be reduced and conservation tillage systems

such as reduced tillage (RT) system can be adopted. Research conducted at CICR, Nagpur indicate that reduced tillage which comprises of pre plant herbicide (dinitroaniline group such as pendimethalin) application, one pass of harrow, two inter row cultivations and one or two hand weeding, is a suitable option for cotton growers of the semi arid tropics of India. A complete elimination of soil disturbance (zero tillage) in the form of inter-row cultivation may not be a feasible technology because of increased weed pressure (Blaise and Ravindran, 2003). In another study, cotton varieties of *G. hirsutum* and the *desi* cottons (G. *arboreum*) were compared with the tillage systems. The Upland cottons were found to be more yielding under the RT systems compared to the conventional till systems (Blaise, 2006). On the other hand, the reverse was true for the *desi* cottons. Probably differences in rooting behaviour contribute to such behavioural response.

With regard to *Bt* cotton hybrids, recent studies indicated that seed cotton yields were greater with the RT than the CT (Blaise, 2011). Averaged over three seasons, yield on the RT plots (1717 to 1740 kg/ha) was significantly higher than CT (1489 kg/ha). On farm trial on farmers fields (Blaise *et al.,* 2005), also indicated the potential of conservation tillage (pendimathalin as preplant (1 kg a.i./ha) and incorporated (0.15 m) + *in situ* green manure with sunhemp (*Crotolaria juncea*) + two inter cultivation and one hand weeding).

Benefits due to RT in cotton may have been achieved due to better weed control. A reduction in weed density, of both monocot and dicot weeds in the RT treatments compared to the CT treatments was reported both on research farm experiments (Blaise and Ravindran, 2003; Blaise, 2006; Blaise, 2011) as well as on-farmers' fields (Blaise *et al.,* 2005). In another long term tillage experiment on soybean, it was clearly demonstrated that the RT and no till systems had fewer grassy weeds than the conventional till systems. Because the conservation tillage systems have crop residue cover that maintains greater soil moisture content and reduces weediness (Bilalis *et al.,* 2003). Apart from the reduced weediness, yield improvements could be due to improved soil structure, soil moisture content. Bordovsky *et al.,* (1994) observed improved yield due to better soil hydro-thermal regimes under conservation till compared to the conventional tillage systems.

Impact on soil properties : Soil structure is impacted by tillage practices with a general agreement that reduced tillage can increase soil organic matter based on research experiments across locations, soils and agro-climates (Bhattacharya *et al.,* 2013; Blaise and Ravindran, 2003; Das *et al.,* 2013; Hulugalle *et al.,* 1997). Thus, soil structure influences crop yield through its complex influence on root-based mechanisms (Passioura *et al.,* 2002). From the studies conducted for more than 10-years at a fixed site on the experimental farm of the Central Institute for Cotton Research, Nagpur, it was observed that the RT treatments had significantly higher proportion of MWD and also the WSA (Table 3).

In the CT treatment, disturbance was greater than 0.10 m (up to 0.20 m in the MB ploughed years and up to 0.10 m in other years). Thus, it is expected that tillage disrupts the soil aggregates. Since the disturbance in the RT plots was restricted to the top 0.05 m, large aggregates tended to increase in the lower soil depth. Consequently, we notice an increase in the MWD and aggregate stability, in terms of WSA. In other

Table 3. Effect of tillage and green manure on aggregate stability and water stable aggregates in the surface 0.05 m soil layer

Treatments	Mean weight diameter (mm)	Water stable aggregates (%)
Tillage systems		
CT	0.43	26.2
RT1	0.47	25.8
RT2	0.47	25.8
LSD (0.05)	NS	NS
Residue management systems		
GM0	0.39	22.4
GM + 100N	0.49	26.3
GM + 80N	0.48	28.0
GM + 60N	0.47	27.1
LSD (0.05)	0.07	2.8

Source: Blaise (2011)

studies, significant improvement in the WSA and MWD were reported in the conservation tillage systems such as reduced and no-tillage. Averaged over tillage systems, residue amended plots had significantly greater MWD and WSA in both the 0-0.05 and 0.05 to 0.15 m soil depths (Table 3). Water productivity amongst the tillage treatments in cotton was 19–27 per cent less in minimum tillage than others tillage treatments. Das *et al.,* (2014) reported improvements in the water productivity in the sandy loams with zero tillage and residue incorporation compared to the conventional tillage systems mainly due to its influence on the soil water retention and absorption.

CONCLUSIONS

The performance of CA in cotton based systems depends on three critical elements-residue generation and its retention, availability of appropriate farm equipments and satisfactory weed management. Non-availability of adequate amount of crop residues, poor efficacy of popular herbicides to manage a wide spectrum of grassy and broad leaved weeds and lack of appropriate from implements for practicing conservation agriculture are the impediments in adopting CA in rainfed cotton system. However, new herbicides for post emergence application have opened up prospects for CA in rainfed areas. Seed cotton yields have been found to increase in the conservation tillage systems as a result of reduction in weed competition and improved soil productivity following addition and conservation of soil carbon. CA will also help mitigate global warming as these systems sequesters C from the atmosphere and stores it in the crop residue and soil and reduces carbon dioxide to the atmosphere. Improved soil productivity is expected to better root growth, increased biodiversity and fewer diseases, and better use of plant nutrients. However, such information is presently limited. Further long-term research is, therefore, needed under various cropping systems and ecological regions for critical assessment of conservation practices.

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Weed management – Present status and future strategies for cotton crop

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ABSTRACT : Weeds are considered as one of the important biotic constraints in agro ecosystems. In many parts of the world, chemical control of weeds with herbicides is the predominant method of weed management not only in cotton but also in major agronomic crops. Weed management in cotton is achieved through a combination of methods that include cultural, mechanical and chemical approaches. As use of herbicides has increased, more and more cases of resistant weeds have been documented. Since the first reported case of weed resistance in 1970, there are 461 unique cases (species x site of action) of herbicide resistant weeds globally, with 247 species (144 dicots and 103 monocots). Weeds have evolved resistance to 22 of the 25 known herbicide sites of action and to 157 different herbicides (Annonymous, 2015). To preserve the utility of herbicides in agriculture, active resistance management is essential, using methods such as herbicide rotation, mixing herbicides with different mechanisms of action, combining non chemical methods like solarization, mulching with organic and inorganic mulches, stale seed bed technique, mechanical removal, growing compatible intercrops as smothering crops and combination of all the above techniques wherever possible keeping in view of the environmental safety and sustainability of the system. Although, herbicide tolerant genetically modified (HTGM) crops offer broad spectrum weed control encouraging the farmers to adopt reduced – or no-tillage cultivation, eliminating the use of some of the environmentally suspect herbicides, there is concern among weed scientists that over-reliance on fewer weed management strategies will result in evolution of resistance to the useful herbicides and/or population shifts to naturally resistant weed species. Recent advances in nano-biotechnology have potential to be explored in developing nano herbicides which can penetrate and kill the weed seeds preventing them from germinating into harmful weeds. The micro encapsulation technology has good potential for controlled release of active ingredient for the extended period of weed control. As the world is gearing up towards challenges of climate change, the weed scientists should initiate and design weed control strategies to meet the challenges posed by weeds keeping the environmental safety at the foremost concern.

Cotton is sensitive to weed competition due to its slow initial growth and wider spacing gives greater chance to high weed infestation. Also, in recent years, *Bt* Cotton which is high yielding and responsive to higher levels of inputs like fertilizers, irrigation etc.,is grown under intensive cropping system, all these factors promote luxurious growth of weeds which grow more quickly than cotton and compete strongly for soil moisture, nutrients, light and space. The yield reduction due to uncontrolled weed infestation was reported between 50-85 per cent (Joshi, 1997). Worldwide consumption of herbicides represents 47.5 per cent of the 2 million tons of pesticide consumed each year(Sopefia *et al.,* 2009).Indiscriminate use of herbicides leads to resistance development. Over the last 40 years, herbicide resistance has increased worldwide at an exponential rate (Heap,2004),mirroring the previous trends observed with insecticide and fungicide resistance (Holt and LeBaron,1990).In parallel,

the rate of discovery of new herbicide modes of action has considerably slowed down (Rasmussen *et al.,* 1999). Management of herbicide resistance is therefore receiving increasing attention to reduce new cases of resistant weed species evolution, in order to prolong the economically useful lives of herbicides. Today, on account of non availability of labour for timely weeding, relatively longer crop duration of cotton crop and enhanced emphasis towards cleaner cotton fibre due to competition from globalization makes weed management as one of the most costly production practices in cotton cultivation and the integration of efficient, economical and environmentally safe weed management system is the need of the hour.

Cotton associated weeds : Weeds of cotton fields vary widely in their floral composition as well as density depending on the ecological situation and crop management. Though about one hundred weed species were reported as associated with cotton, only a dozen of them are responsible for significant yield losses. In India, major weeds associated with cotton crop are *Trianthema portulacastrum,Dactyloctenium aegyptium,Digitaria sanguinalis*,*Ischaneum polosum*,*Alleteropsis crimicina,Eragrostis tenella*, *E.japonica*,*E.major*, *Paspalam disticum*, *Sorghum halepense*, *Brachiaria reptans,Eleusine indica*, *Cyanodan dactylon, Cyperus rotundus*,*Vernonia cinerea*, *Spharamthus indicus*, *Caesulia axillaris*, *Abutilon indica*, *Phyllanthus madraspatensis*, *P.niruri*, *Vicia indica*, *Convolvuilus arvensis*, *Oscimum basilicum*, *Hibiscus malvastrum*, *Lenchus aspewra, Conyza stricta*, *Digera arvensis, Corchorus tridens* and *Enchinochloa colonum*.

Critical period of crop weed competition : The critical period of crop weed competition is the shortest time span in the ontogeny of crop when weeding will result in the highest economic returns. The crop yield obtained by managing weeds during this period should provide crop yields sufficiently close to those obtained by the full season weed control. Balyan *et.al.,* (1983) reported that initial 40 to 60 DAS as critical period of weed competition beyond which keeping the crop free of weeds did not bring any significant improvement in yield. The critical period of weed control was between 20 and 60 DAS as reported by many workers (Shelke and Bhosle, 1989; Saraswat, 1989).The competition of carpet weed (*Trianthema portulacastrum*) was more during initial 50 days after sowing whereas the competition of barnyard grass (*Echinochloa crusgalli*) was more during 50 and 100 days after sowing as inferred by Panwar and Malik (1991) while Douti (1995) specified the critical period of weed competition as 28- 42 DAS for cotton crop. Thus, depending on type of weeds and location, the critical period of weed competition in cotton could be up to 60 days after sowing.

Weed control methods : In India, weed control in cotton has been done traditionally by employing hand labour, animal power or a combination of both. Herbicidal weed control was suggested to be promising. Integrated approach for weed control in cotton reported to be plausible conjecture.

Chemical weed control : Pre emergence herbicides are applied before the crop or weeds have emerged. In annual crops, this is normally done after planting the crop, but before the emergence of weeds. Sprankle (1974) reported that pendimethalin at 0.6 to 1.5 kg/ha incorporated pre sowing gave excellent weed control in cotton. Parshutin *et al.,* (1980)

recommended a pre- emergence application of higher dose of pendimethalin upto 2.0 kg/ha for effective control of weeds compared to trifluralin at 2.0 kg/ha and fluometuron 1.2 kg/ha. Chandler (1984) observed that dinitroaniline herbicides controlled most annual grasses and small seeded broadleaved weeds except perennial weed species. Akhtar *et al.,* (1986) obtained effective control of *kharif* weeds especially broadleaved weeds with pendimethalin 1.5 kg/ha. Panwar *et al.,* (1989) reported that pre-emergence application of pendimethalin 1.25 kg/ha gave the best control of *Trianthema portulacastrum* and *Echinocloa crusgalli.*Solaiappan *et al.,* (1992) observed that pre-emergence application of diethyl ether at 1.5 kg/ha controlled the weeds associated with cotton and enhanced the seed cotton yield. Rout and Satapathy (1998) found that pre emergence application of metolachor at 1.25 kg/ha to cotton controlled weeds efficiently and enhanced the seed cotton yield.

Pre emergence herbicides offer weed control only for a limited period and hence late emerging weeds escape from killing and warrants for post-emergence weed control. Manjunath and Panchal (1989) reported efficient weed control with post-emergence application of fluchloralin 1.5 kg/ha and fluazifop butyl 0.5 kg/ ha. Staple (pyrithiobac-sodium) is a herbicide which has been applied as an over-the top post-emergence herbicide in cotton. It has to be applied at a minimum rate of 0.04 kg a.i./ha to control weed populations from interfering with the cotton harvest (Warrick,1996). Allen *et al.,* (1997) reported that there was no yield reduction with post-emergence application of pyrithiobac at 105 g/ha. Swann and Wilson (2001) reported that pyrithiobac can be applied pre or post emergence to transgenic and non transgenic crop varieties. Directed application of glyphosate can be safely made with shielded nozzles which prevent contact of this herbicide with the crop (Howell and Frans,1980). Best weed control was achieved with post emergence application of glyphosate 1.0 kg/ha as reported by Raja Rajeswari and Charyulu (1996). Glyphosate applied using hooded sprayer provided better weed control (Hawf *et al.,* 1996). Soil application of fluchloralin 1.0 kg followed by foliage spray of glyphosate 0.5 kg at 35 DAS gave the best weed control and best yield in Cotton (Patil *et al.,*1997). Satao *et al.,*(1998 a) reported that the non- selective, non-residual herbicide, Basta (glufosinate ammonium) could be used selectively against weeds in cotton when applied with a hood at 30 DAS and concluded that Basta at 375 g/ha and 450 g/ha was equally effective as paraquat 600 g/ha. Trifloxysulfuron-sodium is a broad-spectrum post emergence herbicide for cotton and sugarcane crop at 7.5 to 15 g/ha and cotton injury was not visibly apparent at six to eight weeks after treatment (Porterfield *et al.,* 2002).

Cotton is a widely spaced crop and hence , the late emerging weeds and perennial weeds that are not being controlled by pre-emergence herbicides can be controlled using nonselective post-emergence herbicides with adequate care using shield or hood so that the herbicide is directed only on weeds without any drift towards cotton crop.

Herbigation : To apply preemergence herbicide on third day of cotton sowing, application of herbicides through drip as herbigation is not advantageous than conventional method due to improper wetting of dry soil under herbigation. However, for post emergence application of residual herbicides, herbigation is found to be the best method than conventional spraying due to thorough wetting

and uniform spread of herbicide molecules under herbigation than under conventional spraying. Hence, for managing weeds under irrigated cotton, herbicide rotation and herbigation are the novel approach and is safe to soil micro flora and succeeding pulse crop (Nalayini *et al.,* 2013).

Integrated weed management : Weed problem is more acute and hazardous due to inadequate field preparation, inefficient weeding during incessant and heavy rains, non availability of labour for timely removal of weeds, inefficient implements etc.,. Therefore an integrated approach of weed management involving herbicides and cultural methods will be effective in controlling weeds. The effect of chemical weed control measure can be strengthened by supplementing with certain mechanical measures and *vice versa.* Herbicides supplemented with an interculture were in a position to enhance the cotton yield by 2 to 2.5 times than either alone (Shelke and Bhosle,1989).Pendimethalin 1.5 kg /ha with one hand hoeing at 45 DAS provided similar yields of seed cotton as that obtained in weed free check (Panwar and Malik,1991).

Chandi *et al.,* (1993) observed that pre-emergence pendimethalin 1.0 to 1.5 kg /ha with one hand weeding 6 weeks after sowing resulted in the highest seed cotton yields. Fluchloralin, pendimethalin, butachlor and diuron as pre-emergence spray followed by interculture and hand weeding recorded higher *kapas* yield over sole application of paraquat and glyphosate as post-emergence spray (Raja Rajeswari and Charryulu,1996). At hilly zone of Karnataka, clomozone 1.5 kg a.i./ha + hand weeding was the best treatment in controlling weeds and produced higher seed cotton yield (Madiwaler and Prabhakar,1998).Glyphosate at 1.0 kg as directed spray on 20 DAS using hood followed by one hand weeding at 45 DAS recorded the lowest weed DMP on 60 DAS (Nalayini *et al.,*1999 a) and nutrient uptake by weeds on 90 DAS (Nalayini *et al.,*1999 b).Highest seed cotton yield with Pendimethalin 1.5 kg as pre-emergence followed by one hoeing as reported by (Brar *et al.,*1999).According to Sreenivas (2000) alachlor at 2 kg/ha and diuron at 0.75 kg/ha plus glyphosate at 1.5 kg/ha recorded better weed control in cotton. Pendimethalin 1.0 kg as pre emergence herbicide followed by one hand weeding at 35- 40 DAS and mixture of pyrithiobac sodium 50g + quizalofop ethyl 50 g on 60 DAS was found to be efficient and more economical for managing weeds of irrigated cotton (Nalayini *et al.,* 2012)

Stale seed bed technique : A stale seed bed is a seedbed which has been prepared and given a false start some weeks before the seed is due to be sown, any weed seeds in the bed will be encouraged to emerge and grow so that they can be raked out and killed before the actual cotton crop is sown. This technique reduces the number of weeds which have to be controlled when the cotton seedlings start to grow in the field. Stale seedbeds are established several weeks or months before planting (Hydrick and Shaw (1994), Minton *et. al.*, (1989). Stale Seed Bed Technique (SSBT) using a mixture of pendimethalin 1.0 kg.a.i + glyphosate 1.0 kg.a.i./ ha one week after pre sowing irrigation (one week before sowing) recorded the highest weed control efficiency of 85.2 per cent on 35DAS (Nalayini and Suveetha (2012).

A perusal of the above literatures suggests that integrated approach is the most effective approach for controlling weeds in cotton.

Biotechnology applications in chemical weed management

Herbicide tolerant genetically modified crops : Recently, genetically modified crop varieties with two biotech traits (stacked trait crops) have been made commercially available and currently cultivated in several countries. Stacked trait products are mainly represented by plants that have been genetically modified to exhibit tolerance to glyphosate or glufosinate and resistance to lepidopteran pests. Before the emergence of plant genetic engineering, option for selective crop protection against herbicides was limited. Development in plant genetic engineering and knowledge of biochemical action of herbicides on plants spurred innovative approaches to engineer crops to withstand herbicides. These strategies usually involve isolation and introduction of a gene from other organisms, mostly bacteria which is able to overcome the herbicide induced metabolic blockage. Tolerance to herbicide glufosinate (Basta ^{R})is conferred by the bacterial gene bar, which metabolizes the herbicide into non toxic compound (Thompson *et al.,*1987). Glyphosate resistance is achieved by the introduction of either Agrobacterium gene from CP_4 that codes for a glyphosate insensitive version of the plant enzyme,EPSP Synthase or gox gene from *Achromobacter,* which codes for a glyphosate oxireductase in the breakdown of glyphosate . A number of other genes have been identified that can alleviate the herbicide action through various ways (such as detoxification, sequestration etc.,) and thus confer resistance to the plants carrying them. Thus genetic engineering technology has made it possible to tailor crop varieties to resist specific herbicides by introducing relevant genes. HTGM crops will allow farmers total control of weeds. HTGM crops are gaining farmers' acceptance because of several advantages such as increased flexibility to manage problem weeds, prevention of multiple use of herbicides, reduction in total herbicide use, greater adoption of conservation tillage, less herbicide carry over etc.,

Concerns and apprehensions of HTGM crops : The use of herbicide resistant crops undermines the possibilities of crop diversification, thus reducing agro biodiversity in time and space (Altieri,1994).Effects of introduction of herbicide resistance crops on biodiversity also reported by (Amman,2005), escape of transgenes from HTGM crops, non selective herbicides may wipe out all vegetation except the HTGM crops, development of herbicide resistance in weeds, shift in weed flora etc., There is potential for herbicide resistant varieties to become weeds in other crops (Holt and Le baron,1990). The HTGM crops may replace labour and deny rural woman the livelihood as most of the weeding is done by them (Varshney and Naidu,2009).Also in countries like India where multiple crops are grown such as under intercropping system wherein compatible intercrops are grown with cotton, the use of HTGM technology is not possible. *Bt* resistance and herbicide resistance are qualitatively different. If insects develop resistance against a particular *Bt* toxin, alternative *Bt* toxin with different modes of action or target sites can be deployed. Various strategies such as pyramiding of different *Bt* genes and maintaining refugia have been suggested to delay development of resistant insects. Similarly, strategies have been defined to delay development of herbicide resistance weeds in the case of conventional crop varieties. These include, combined or sequential use of herbicides with different mode of action, crop rotation, integrated weed control etc., (Das

and Duary,1999) and combining non chemical approaches like crop rotation, cultivation, mulching with organic and inorganic materials and solarization helps in managing herbicide resistance development (Timothy et al 2000).In the case of genetically engineered HTGM varieties, these strategies are less relevant. When the herbicide could be applied at various stages of crop growth, farmers may not opt for integrated weed control measures. Similarly, when different crops carry engineered resistance to the same herbicide, use of different herbicides may not remain an option .Once the weeds develop resistance, through either acquisition of the gene from the HR variety or by mutation, they will remain resistant against the herbicide. Replacement of the herbicide is the only option in such a scenario. Since development of new and safer herbicides is time and resource demanding, development of new herbicides is not likely to keep pace with emergence of HR weeds. A major current concern with the introduction of glyphosate or glyufosinate resistant transgenic crops is that if the weeds develop resistant, these environmentally benign herbicides will become ineffective and will force use of other less desirable herbicides for weed control. Already, glyphosate resistant *Lolium* populations have emerged in Australia (Powels *et al.,* 1998), USA (Simarmata *et al.,* 2001) and South Africa (Cairns *et al.,* 2001).Further, glyphosate resistance has also been recorded in *Eleusine indica* in Malaysia (Lee *et al.,*1999; Tran *et al.,* (1999), *Lolium multiflorum* in Chile (Perez and Kogan, 2003) and *Conyza Canadensis* (Van Gessel, 2001 ; Mueller *et al.,* 2003)

Thus as technology, herbicide resistant crops offers opportunity for efficient control of weeds. However, doubts remain about the long term viability of this strategy, especially the emergence of herbicide- resistant weeds following wide spread cultivation of HRGM crops and the best herbicides may not be available even for conventional weed control (Bhat and Chopra,2006).

Nano Agrobiotechnology and weed management : Systems of Controlled liberation (SCL) represent an alternative to the conventional systems of herbicide application. This process is defined as " a technique or method where the active agent is available for a specific product to a speed and duration designed to achieve the intended effect (Scher,1999).The herbicide SCL is a technology wherein an active ingredient is available for a specific goal at a concentration and with a duration designed to achieve the intended effect, aiming to reach optional biological effectiveness and to reduce any harmful effects (Ruegg *et al.,*2007 : Undabeytia *et al.,* 2004 : Fernandez – Perez, 2007). Reducing herbicide levels also reduces costs for farmers as well as for companies (Markus,1996).Reduction in volatilization loss of applied herbicides due to micro encapsulation has been reported thus diminishing the presence of herbicide in the atmosphere (Whienhold and Gish, 1994 : Dailey, 2004) , reduction in phyto toxicity in crops due to micro encapsulation has also been reported (Bernards *et al.,*2006).Micro-encapsulation has been proven to improve pesticide effectiveness in comparison to commercial formulations (Greene at al., 1992 ; Vasilakoglou and Eleftherohorinos, 2003 ; Hatzinikolau *et al.,*2004, Sopefia *et al.,* 2007, 2008). Development of nanoherbicide to penetrate cells is under development jointly by Agricultural Rresearch Institute in Mexico and India that would attack a weed's seed coating ,germination of weeds would be prevented and the seed would thus be destroyed even when it

is buried deep in soil, below the reach of tillers and conventional herbicides because soil particles would not be able to stop the minute herbicide nanoparticle from migrating down.Breaking the dormancy of Cyperus rotundus usng zinc oxide nanoparticles at 3g/ kg of tubers for the management of wCyoerus rotundus has been reported (Viji and Chinnamuthu, 2015).

The perusal of the literatures on micro encapsulation of herbicides show that in future controlling weeds with micro encapsulated herbicides will be the potential technology to achieve season long weed control with lesser costs and risks to the environment.

Introgression of crop genes and trangenes into weeds : Gene flow between transgenic crops and conventional varieties or their wild relatives has been cited as one of the central ecological risks associated with the application of biotechnology to crop production (NRC, 2000). Primary concerns of ecologists have been that genes conferring insect resistance might increase ûtness, competitive ability, and invasiveness of the crop itself, or cause increased invasiveness of wild crop relatives that may obtain the trait through hybridization and subsequent gene ûow (Kareiva *et al.,*1994, Hails, 2000).Assessing gene flow consequences is challenging, because it is difficult to predict the ecological effects of transgenes that are integrated into different genetic backgrounds or expressed in different ecological contexts. Conner *et al.,* (2003) proposed three potential consequences of gene flow from GM crops: "transfer" of the genes to nearby crops causing the trait to spread (intentionally or unintentionally); "escape" of the insect resistance genes to non-cultivated related species and increase in the potential of insect-resistant versions of crops to become established as weeds.

In cotton, the risk of transfer or escape of the *Bt* gene from *Gossypium hirsutum* to the sexually compatible species could not be ignored. The potential transfer through gene flow from herbicide resistant crops to wild or semi domesticated relatives can lead to the creation of superweeds (Lutman.1999).The weed relatives of Cotton that have been reported are *Gossypium tomentosum* which is found in Hawaii and in India, *Gossypium stocksii* has been reported .Crosses among tetraploid *Gossypium* species can be successful, while crosses between tetraploid and diploid *Gossypium* species are essentially unknown without human intervention (Brubaker *et al.,* 1999).A herbicide resistance trangene alone confers no fitness advantage in areas where the herbicide is not sprayed. If it is transferred from the crop to a related weed species,the biggest concern is for the farmer who must cope with the herbicide resistant weed. An herbicide resistance trangene in a crop can greatly increase the chance of survival of interspecies crosses by eliminating competition of other herbicide susceptible weeds (Keeler *et al.,*1996).If the crop also contains trangenes conferring other survival – enhancing traits, such as resistance to insects and/or pathogens, the resulting cross and further back crosses with the weedy parental species might confer enhanced fitness outside the agricultural setting, resulting in ecological disruption.

Glyphosate and glufosinate – ammonium on soil : Glyphosate and glufosinate herbicides are widely used in weed management and the importance of these two herbicides has increased in the last years due to the increased demand for herbicide – tolerant crops (Shaner,2000). Crops tolerant to these herbicides

are the most cultivated genetically modified crops and environmental issues concerning the cultivation of crops tolerant to these herbicides are of current interest (Engel et al ., 2002).Glyphosate and glufosinate are non residual herbicides that degrade readily from soil with estimated half-lives ranging from 7 to 60 days and from 1 to 25 days respectively (Giesy *et al.,*2000). Degradation by soil micro-organism is the predominant way by which these herbicides are metabolized in soil (Tebbe and Reber,1988 ; Giesy *et al.,* 2000). No detrimental effects were observed on soil microbial activity and biomass when glyphosate and glufosinate were applied at normal agricultural rates under laboratory and field conditions (Wardle and Parkinson,1992: Accinelli *et al.,* 2002). According to Wauchope *et al.,* (2001) glyphosate and glyfosinate replace herbicides that are in general more persistent in soil and absorbs less to soil particles.

Herbicides and microorganisms : Intensive cultivation necessarily employs chemical herbicides for effective control of various weeds. In recent days, contamination of environment by these toxic xenobiotics on environment has raised serious concerns all over the world. Despite these concerns newer herbicide molecules are being added regularly. Therefore, issues concerning degradation, detoxification of applied herbicides and use of eco-friendly weed management strategies will receive greater attention now than ever before. Apart from causing pollution, the applied herbicides also have an impact on non target soil microorganisms. Hence, impact of herbicides on soil microorganisms should be an important consideration for employing herbicide in weed management. Strategies for effective weed management must take into account the effect of herbicides on soil borne pathogens,plant growth promoting microorganisms and their saprophytic survival. The involvement of microbial endophytes of weeds need thorough investigation as their role in conferring protection and ecological fitness in weeds has been demonstrated conclusively. Identification and development of native microorganisms with ability to degrade weed seeds to kill the weeds either directly or indirectly or by production of secondary metabolites with herbicidal properties will have relevance in eco-friendly weed management strategies (Patil *et al.,* 2009).

Impact of climate change on weeds and herbicidal weed management

Enhanced CO_2 on weeds : Weeds have a greater genetic diversity than crops. Consequently, if a resource (light, water, nutrients or carbon dioxide) changes within the environment, it is more likely that weeds will show greater growth and reproductive response. It can be argued that many weed species have the C_4 photosynthetic pathway and therefore will show a smaller response to atmospheric CO_2 relative to C_3 crops. However, this argument does not consider the range of available C_3 and C_4 weeds present in any agronomic environment.Today,for all weed/crop competition studies where the photosynthetic pathway is the same, weed growth is favoured as CO_2 is increased. There are some studies (Ziska and Teasdale 2000; Ziska *et al.,* 2004) that demonstrates a decline in chemical efficacy with raising CO_2 .Dilution effect of glyphosate for controlling Canada thistle under elevated CO_2 has been reported (Ziska *et al.,* 2004).Biological control of pests by natural or manipulated means is likely to be affected by increasing atmospheric CO_2 and climate change. Climate as well as CO_2 could alter the efficacy of weed bio control agents

by potentially altering the development, morphology and reproduction of the target pest.

Temperature increase on weeds and weed management : Increasing temperature may mean an expression of weeds into higher latitudes or higher altitudes.. With an increase of 3^0 C ,many weeds which are not problematic today may become aggressive as reported for itch grass (Patterson,1995).Weeds being major pests competing for basic inputs with crop and being aggressive have developed all strategies to combat change in climate than crop (Devendra *et al.,* 2009).Witch weed, a root parasite of corn, is limited at this time to the coastal plain of North and South Carolina and with an increase of temperature of 3^0 C, it is speculated that this parasite could become established in the Corn Belt with disastrous consequences. Clearly any direct or indirect impacts from a changing climate will have a significant effect on chemical management. Changes in temperature, wind speed, soil moisture and atmospheric humidity can influence the effectiveness of applications. For example, drought can result in thicker cuticle development or increased leaf pubescence, with subsequent reduction in herbicide entry into the leaf and hence suitable herbicide formulations are to be standardized to make the herbicide enter into the target sites.

The literatures on impact of climate change on weeds clearly suggest that many weeds which are less problematic or sleeper weeds may become aggressive and troublesome weeds due to climatic change and also the weeds may extend their spread to newer areas and hence it is necessary to generate information on these crucial aspects in order to equip ourselves to meet any challenges in weed management due to climate change.

CONCLUSION

In Cotton, The greatest competition from weeds usually occurs early in the growing season. However to harvest clean and uncontaminated fibre, it is essential to keep weed free up to harvest. The mechanical removal of weeds is tedious and costly. Although, the pre-emergence or pre plant incorporated herbicides take care of weeds during early days, the late emerging weeds are to be controlled by mechanical or directed application as we do not have selective and safe herbicides. Micro encapsulated nano herbicide formulations have potential in future weed control programme. In most major crops, stacked genes technology widen the options for efficient and economical pest management and strongly impacting weed management choices in short to medium term. However, in long term, the problems due to herbicide resistance and super weeds as a result of this technology will be a serious threat if the traditional resistance management strategies like, crop rotation, cultural methods, non chemical approaches and herbicide rotation are not followed. However, HTGM crops offer farmer a powerful new tool that can be incorporated into an integrated pest management strategy which can be used for many years for managing weeds economically and effectively. In future, nanotechnology offers opportunity and potential in scientific weed management.

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Does *Bt* cotton differ from conventional cotton on agronomic requirement ?- A Review

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Cotton, being the major fibre crop in India and world, has exercised a profound influence on man and matters. Amongst the cotton growing countries, India occupies the foremost position in terms of its acreage, extending over 12.7 mi ha with production of around 373.9 lakh bales. Before the *Bt* cotton era the excessive and indiscriminate use of pesticides had caused insecticide resistance in major pests like bollworms and emergence of secondary pests in epidemic form making the cotton production risk prone compared to other crops (Kranthi *et al.,* 2002). *Bt* cotton has inbuilt resistance of bollworm due to the *Bt* gene, thus protect early formed bolls and kept intact in the plant. *Bt* cotton is gaining popularity with farmers because of effective control of bollworm complex besides higher productivity (Fitt *et al.,* 1994; Flint *et al.,* 1995; Harris *et al.,* 1996) and ultimately resulted an expansion of area under *Bt* cotton in India from 38,000 ha in 2002 to 11.6 Mio ha in 2014 (Clive 2014).

Agronomic performance of *Bt* transgenic cultivars may vary substantially from their non transgenic counterparts. The expression of transgene can be influenced by transgene x genetic background effects (Sachs *et al.,* 1998) and the transformation techniques randomly incorporate a gene into the host (Altman *et al.,* 1991). When a transgene is introgressed into an elite genetic background, the agronomic performance may be altered because of all the donor DNA from the originally transformed line is not eliminated through back crossing (Falconer, 1989). Introduction of the *Bt* gene altered the morphological, phenological and physiological characteristics of the introgressed cultivars as reported by Venugopalan *et al.,* 2009.

Bt cotton hybrids are compact and relatively early maturing than isogenic non *Bt*. The *Bt* hybrids have an optimum number of functional leaves and are more efficient in converting assimilates to the cotton bolls. As a result, *Bt* cotton hybrids retain more bolls/plant and simultaneously with less boll locule damage resulting in an overall yield advantage. Chen *et al.,* (2012) reported that the *Bt* isogenic line had higher rate of effective bolls. The *Bt* isogenic line had higher Chl a/b, soluble protein, P and Cu at boll setting stage (BSS) and . at initiation of flowering stage (IFS) had significantly higher concentrations of Ca, Mg, Cu, Zn, Mn and Fe, whereas it had lower in concentrations of P, K and B at IFS, and K, S, Zn and Fe at BSS. Wude Yang *et al.,* (2012) that the concentration of the *Bt* protein in the rhizosphere soil reached a peak at 56.14 ng g"1 during the flowering period. However, the *Bt* protein would not continuously accumulate in the soil. The rhizosphere soil of was more suitable for the growth and proliferation of bacteria and fungi but it had no significant impact on the number of actinomycetes.

Additionally *Bt* cotton varying from non *Bt* with respect to retention of early formed bolls, synchronous bursting, lower bad *kapas* content

and less labour required for harvest. To utilise the full potential of the *Bt* technology, the advantages associated with these hybrids have to be exploited by modifying or refining the existing agronomic practices to suit different cotton production domains, besides managing limitation like reddening and pre mature senescence associated with *Bt* cotton hybrids.

Compact growth : Growth and development has a direct bearing on reproductive efficiency and seed cotton yield. *Bt* hybrids recorded 6.95per cent less plant height, 10.81per cent less leaf area index (LAI) than non *Bt* hybrids (Rekha, 2007). Plant height and dry matter accumulation, by the Bunny *Bt* was found to be significantly lower , while number of bolls and seed cotton yield were significantly higher than Bunny non-*Bt* (Sunitha et al .,2010).Root volume was significantly higher in *Bt* than non-*Bt* isogenic lines (Sarkar *et al.,* 2008). *Bt* recorded a mean of 25.1 bolls, while it was only 8.8 in non-*Bt* cotton hybrids. Numerically 18.5per cent higher number of squares was observed in *Bt* hybrids. At Coimbatore, non-*Bt* hybrids recorded 40per cent higher LAI and increased plant heights than *Bt* hybrids under rainfed condition (Sankaranarayanan *et al.,* 2011). The *Bt* hybrids were short statured as reported by Mayee *et al.,* 2004. Further study at Nagpur and Coimbatore revealed that less dry matter production, shorter in stature with lesser LAI in *Bt* hybrid as compared to non *Bt* counterpart (Anonymous, 2002).

Compact growth of *Bt* hybrids required reduced spacing and reduction of 15 cm from existing spacing, realized yield advantage of 2.5 q /ha with *Bt* hybrid under front line demonstrations conducted at Chandrapur and Yeotmal districts of Maharashtra with boll guard II during 2007 and 2008.

Apparently from the above observations, the dwarfness in *Bt* might have resulted due to individual or combined effect of transgene x genetic x environment interaction or utilization of more nutrient energy (sink strength) for the nourishments of (maximum) of bolls that were free from bollworm damage due to active expression of *Bt* toxin. In other words, retaining of early formed bolls, prevented from attack by bollworms as a result of inbuilt protection mechanism or individual or combined effect of transgene x genetic x environment had suppressed the vegetative growth parameters including plant height, LAI and monopodia branching but at the same time harnessed the reproductive parts especially developing squares and harvestable maturing bolls in *Bt* cotton. Non-*Bt* hybrid, because of absence of *Bt* gene, was susceptible to bollworm damage and frequently losing the early formed square and bolls and therefore has been induced to grow vegetatively to make compensation for the lost squares and bolls by further branching and axils production to accommodate new squares. Thus, the unique nature of the *Bt* hybrids capable of retaining early produced bolls therefore, shows limited canopy growth and thus occupies less land when compared to non-*Bt* hybrids, keeping the land mass unutilized and offers chance for growth of weeds. Therefore, alterations in row and plant to plant spacing is much essential to better utilize the land and offers the chance for enhanced production and productivity of cotton. The space may also be utilized for accommodating higher population or intercropping tactics also, a practice that enhances the farmers' income base besides a better pest management tool too.

Early maturity : Inbuilt resistance to bollworm complex leads to retention of early formed fruiting parts that might have

pronounced earliness in *Bt* cotton hybrids (Anonymous 2002; Mayee and Rao 2002, Venugopal *et al.,* 2004) and at least 20-30 days early maturity advantage is prevalent with *Bt* cotton hybrids than their non-*Bt* counterparts (Mayee *et al.,* 2004).Higher sink in *Bt* cotton leads to lower source to sink ratio, faster senescence and crop maturity compared to the non *Bt* version (Hebbar *et al.,*2007). Sometimes, the increased assimilate demand of early high fruit retention reduces the resources for continued growth and fruiting, leading to early maturity and reduced yields (Bange *et al.,* 2008).Rekha (2007) observed that *Bt* hybrids matured five days early compared to non-*Bt* hybrids. Bartlett's earliness index of 0.80 with *Bt* hybrids when compared to 0.67 for conventional hybrids was observed (Deosarkar, 2004) (Table 1) and also in another study a 10per cent increase in earliness index of *Bt* hybrids (0.7) than non-*Bt* hybrids (0.64) is reported (Sankaranarayanan *et al.,*2011b*)* as influenced by the early boll setting and boll opening . These studies concluded that *Bt* is maturing earlier than non *Bt*. This earliness associated with *Bt* hybrid could be exploited under cotton based cropping system for timely sowing of succeeding winter wheat and other crops in North and Central zones of India, rice in south zone and also to achieve yield maximization through rotational crops.

Agronomy practices : The yield losses is due to climatic aberrations are higher in cotton (60per cent) as compared to other crops like cereals, oilseeds and pulses (30per cent) (Dason, 1996). The high sensitivity of *Bt* hybrids to leaf reddening induced by drought and low temperature indicates that *Bt* cotton hybrids are more influenced by climate.

Soil : In India, *Bt* cotton is grown in varied soils from deep heavy vertisol to sandy loam alluvium soil. In areas like Maharastra and Madya Pradesh, *Bt* cotton is grown on large scale in shallow black soil with low fertility levels and moisture retaining capacity. In the south zone, *Bt* cotton is grown in hot semi arid regions, both under rainfed and irrigated conditions in

Table 1. Yield and earliness index of *Bt* and non *Bt* hybrids

Non *Bt* hybrids	Yield (kg/ha)	Earliness index	*Bt* hybrids	Yield (kg/ha)	Earliness index
Akka non *Bt*	1545	0.57	Akka *Bt*	2681	0.78
Ankur 2534 non *Bt*	1105	0.73	Ankur 2534 *Bt*	1561	0.82
Bunny non *Bt*	1879	0.73	Bunny *Bt*	2221	0.84
Ankur 2226 non *Bt*	716	0.67	Ankur 2226 *Bt*	1325	0.76
Mean non *Bt*	1331	0.68	Mean *Bt*	1947	0.80

Source: Adapted from Deosarkar (2004)

Table 2. Seed cotton yield of MECH 162 *Bt* hybrid under different situations*

Genotypes	Deep soil +HR	Deep soil + LR	Medium soil + HR	Medium soil + LR	Shallow soil + HR	Shallow soil + LR
MOL 2463	1139	1120	1207	879	1244	802
NHH 44	1058	1500	1107	840	1055	612
MECH 162 *Bt*	1263	1580u	1340	1259	1655	1347

Bhatade *et aI.,* (2006) *High rainfall=HR and Low rainfall=LR

Table 3. Influence of soil depth on the yield of *Bt* and non *Bt* hybrids

Parameters	MECH 184 *Bt*	MECH 184 non *Bt*	MECH 162 *Bt*	MECH 162non *Bt*	MECH 12 *Bt*	MECH 12 non *Bt*	NHH44
Shallow	18.25	13.6	18.53	16.0	11.75	15.2	12.8
Medium deep	21.14	14.18	21.26	14.35	17.7	14.1	11.25

Jagvir Singh *et aI.,* (2004)

medium black soil, red and black soil and coastal alluviums. While assessing the performance *Bt* hybrid in deep, medium and shallow soils combined with high and low rainfall under Marathwada region, it was reported that *Bt* hybrids performed better (Table 2) than non *Bt* *hirsutum* hybrid and *arboreum* variety in all situations (Bhatade *et al.,* 2006).

Better performance of *Bt* hybrids, MECH 184, MECH 162 and MECH 12 was noticed compared to non *Bt* hybrids in medium deep than shallow soil (Jagvir Singh *et al.,* 2004) with a higher harvest index of MECH 184 *Bt* (46.5per cent) and (51.5per cent) on shallow and deep soil respectively than MECH 184 non *Bt* (Table 3).

Sankaranarayanan and Nalayini (2015) reported that, *Bt* hybrids (1691 kg/ ha) produced higher seed cotton yield than non-*Bt* hybrids (1092 kg/ ha), while the controlled variety (LRA 5166) performed the average of these two (1399 kg/ ha).

Method of planting : In accordance with conditions laid down in the Genetic Engineering Approval Committee (GEAC) *Bt* cotton should be planted in the centre of the plot surrounded by non-*Bt* isogenic lines at the border in five rows for every acre of planting area. The size of refuge (the non *Bt* belt surrounded by *Bt*) should take at least five rows of non *Bt* or 20per cent of the total land sown, whichever is more . To meet the requirement of non *Bt* refuge (corresponding non *Bt* or non *Bt* of equivalent fibre quality) , seeds are provided with *Bt* seeds (450g of *Bt* + 120 g of non *Bt*).

For yield miaximization, following of proper seed rate and spacing are essential. Depending upon the hybrids and soil fertility, spacing varies, which ultimately decide seed rate of the hybrid. In sowing of *Bt* cotton, single seed is dibbled at a depth of 3 cm in the furrows, where fertilizers and insecticides are applied under irrigated condition. However, planting of single seed with pulses/castor/sunhemp seed facilitate the germination of cotton vigorously and later the extra plant is to be clipped to avoid root injury of cotton seedlings . In the Yellow River valley, China, transplanted *Bt* hybrid cotton yielded 31per cent more than the direct seeded cotton (Dong *et al.,* 2005). Leaf reddening in transplanted *Bt* cotton (20-25 days old) was reduced (7.8 to 9.2per cent) compared to dibbled cotton (19per cent).

Time of sowing : Optimum time of sowing is varying depends upon the climate, hybrids, rainfed and irrigated condition across the country. Sowing of conventional cotton beyond the recommended dates of planting invites the incidence of pests especially bollworms complex in many parts of the country, besides exposing the crop to unfavourable climatic conditions during crop maturity phase. As the *Bt* cotton is effective against bollworm complex and shows early maturity, delay in

sowing of *Bt* cotton may not much aggravate the bollworm incidences or predispose them to unfavourable climatic conditions and thereby ensuring normal yields. The experiment on time of sowing of *Bt* hybrids conducted in different centres of AICCIP project revealed that *Bt* cotton following of the timely sowing recorded 1693 kg/ha as compared to delay in sowing of 1389 kg/ha at Surat, whereas delayed sowing increased the seed cotton yield in *Bt* cotton hybrids and at the same time reduced in non *Bt* hybrids. Prakash *et al.,* (2008) observed that a 10 day delay in sowing time (from the optimum date of 12 August) had no effect on seed cotton yield of Bunny *Bt* and RCH 2 *Bt* in the winter irrigated tract of southern zone. In contrary to that reports from All India Coordinated Cotton Improvement Project (AICCIP) indicate that a 20 day delay in sowing caused a reduction in the yield of *Bt* cotton by 18per cent at Surat, Gujarat and 22per cent at Khandwa, Madhya Pradesh (Singh *et al.,* 2008) and 31per cent at Dharwar, Karnataka (AICCIP, 2009).

Plant Population : The success in increasing the *Bt* cotton productivity depends on adoption of improved agro techniques. Plant population (spacing) contribute higher towards maximizing the yield. Optimum plant density is depending on the inherent vegetative habit of variety/hybrid and conditions of soil fertility, moisture and cultural practices (Bapna *et al.,* 1976). Similarly, optimum row spacing have a bearing on seed cotton yield. An yield of 2300 kg/ha was observed at Coimbatore, Tamil Nadu with closer planting at 75 x 60 cm (22,222 plants/ha) (Sankaranarayanan *et al.,* 2011a). Narrow spacing of 67.5 x 60 cm recorded significantly higher seed cotton yield over wider spacing (100 x 60 cm) in canal command areas of northwestern Rajasthan (Nehra *et al.,* 2004). Raghuramireddy *et al.,* (2007) reported that under 90 cm row spacing, closer intra-row spacing of 30 cm (3111kg/ha) and 60 cm (3019 kg/ha) significantly enhanced seed cotton yield over 90 cm (2761 kg/ha). The highest seed cotton yield of RCH-134 *Bt* cotton hybrid was recorded at 67.5 x 90 cm spacing by Butter and Singh (2006) at Bhatinda. Vishwanath (2007) recorded significantly higher seed cotton yield with 90 x 30 cm spacing (2479 kg /ha) as compared to control (2139 kg /ha). The highest seed cotton yield recorded at 90 x 45 cm spacing for RCH-2 *Bt* cotton under rainfed conditions as reported by Bhalerao et al (2010). MECH 162 and RCH 2 *Bt* hybrids adopted at a spacing of 90x60 cm had recorded significantly higher seed cotton yield as recorded by Kalaichelvi(2009). Aruna and Sahadeva Reddy (2009) confirmed that planting *Bt* cotton at 90 cm x 45 cm gave higher kapas yield in scarce rain fall zone of Andra Pradesh. Manjunatha *et al.,* (2010) reported spacing of 60 x 30 cm recorded significantly higher (21.11 q / ha) seed cotton yield than wider spacing of 90 x 60 cm (15.59q /ha), 75 x 30 cm (18.85 q /ha) . The plant geometry of 120 cm × 45 cm recorded highest seed cotton yield (26.46 q/ha) and stood significantly superior over other planting geometries. It has 8.9 and 13.5per cent yield increment over 90 cm × 60 cm and 180 cm × 30 cm, respectively(Dadgale et al 2014).

The above said experimental results on spacing requirement of *Bt* hybrids may be generalized that *Bt* hybrids plant occupies less space because of compact growth in habit. The compact growth of *Bt* hybrids is provided opportunity to keep more population per unit area as compared to non *Bt* to reach optimum and higher plant population per unit area ultimately result in yield increase.

Nutrient management : Fertilizer is the

foremost input, towards yield maximization. Application of 150 per cent of RDF (135: 67.5:67.5 NPK (on par with 125 per cent RDF) showed significantly higher number of bolls, single plant yield, and seed cotton yield (Sankaranarayanan, *et al.,* 2011a) . Significant differences were not observed in seed cotton yield in the first picking but in the second picking, 125 per cent of recommended dose of fertilizer (RDF) resulted in significantly higher yield as compared to 100 per cent RDF (Jagvir *et al.,* 2000). Vishwanath (2007) recorded significantly higher seed cotton yield 150 per cent RDF (2420 kg /ha) as compared to control (2139 kg /ha). MECH 162 and RCH 2 *Bt* hybrids applied with fertilizer levels of 160:80:80 kg of N, P and K/ha had recorded significantly higher seed cotton yield as compared to recommended level of 120:60:60 kg of N,PandK/ha (Kalaichelvi, 2009). Hallikeri *et al.,* (2011) indicated that increasing level of nitrogen from 80 to 120 and further to 160 kg/ ha significantly increased seed cotton yield by 12 and 19per cent,respectively. In contrary to that of higher fertilizer requirement and response of *Bt* hybrids observed in many field trials, non significant differences of seed cotton yield observed under different fertility levels by Raghuramireddy *et al.,*(2007). *Bt* hybrids performed better than conventional hybrids and required same quantity of fertilizer as observed by Hallikeri *et al.,*(2004).

Rajendran *et al.,* (2009) found that application of 150 per cent RDF combined with TNAU MN mixture recorded 26 per cent higher seed cotton yield in *Bt* cotton. On vertic Ustropepts of Coimbatore, Bandhopadhyay *et al.,* (2009) observed 60 kg N/ha as optimum dose for high yield and was adequate for conventional hybrids But *Bt* cottons are slightly more responsive to N application . Moreover, *Bt* cotton had a higher N content than non-*Bt* cotton suggesting that they may have a greater N uptake and metabolism than non-*Bt* cotton (Showalter *et al.,* 2009). Nehra and Yadav (2011) concluded that 108 X 60 cm spacing with application of 100per cent RDF (150 kg N and 40 kg P2O5 /ha) seems to be the optimum dose for RCH 134 *Bt* cotton hybrid in Canal command area of North-west Rajasthan.Late application of N and K fertilizer delay the senescence, (Dong *et al.,* 2005) . Hosmath et al (2014) Foliar application of KNO3 2per cent, soil and foliar application of MgSO4 @ 25 kg/ha and 1per cent respectively, and soil application of MgSO4 @ 25 kg/ha alone increased the seed-cotton yield by 25.0, 24.4 and 24.3per cent respectively, over the RDF. The yield of *Bt* hybrids (MCEH 184 and RCH 2) was higher than non-*Bt* NCS 145 and among *Bt* hybrids, the medium duration RCH 32 *Bt* was superior to short-duration MECH 184 under normal sowing date (Venugopalan et al,2012). *Bt* protein had some inhibitory effects on alkaline phosphatase activity in the rhizosphere soil, and it might promote dehydrogenase activity during the blooming period. However, it had no significant influence on protease, urease, or sucrose activities.and had no significant impact on the contents of organic matter, total nitrogen, available nitrogen, or potassium in rhizosphere soil. It could significantly decrease the content of available phosphorus during the flowering period(Wude yang et al (2012) .

Use of organic inputs is a long term remedy in order to get rid of any adverse effect of intensive agricultural system and to restore the soil health and environment. Performance of *Bt* hybrids under different organics revealed that cotton yield (kg/ha) was significantly higher in *Bt* (1172) over DHH-11 (876) and non *Bt* (719). *Bt* cotton hybrids had not influenced significantly on soil pH, EC, available N,P and K status in

comparison to non *Bt* and locally adopted genotypes (Sankaranarayanan *et al.,* 2008).The similar results were reported by Hosmath *et al.,*(2004a) except for less availability of nitrogen (Table 4). Blaise (2011) reported that N100, GM + N100 and GM + N80 (1687–1734 kg/ ha) did not differ and were significantly better than the GM + N60 (1303 kg/ ha).

Water management : Water requirement of *Bt* cotton hybrids ranges from 60 to 120 cm in different regions. Early maturity

Table 4. Yield, soil properties and available nutrient status in *Bt* and non *Bt* hybrids

Parameters	*Bt*	Non *Bt*	DHH 11	S.E	C.D(0.05)
Seed cotton yield (Kg/ha)cotton yield	1172	719	876	27.0	106
Organic carbon (%)	0.66	0.67	0.65	0.02	NS
EC(dS/m)	0.27	0.28	0.27	0.01	NS
pH	7.94	7.98	7.98	0.13	NS
Nitrogen (Kg/ha)	276.7	214.3	280.9	7.12	27.9
Phosphorus (Kg/ha)	21.4	20.4	20.82	0.6	NS
Potassium (Kq/ha)	333.5	330.7	329.8	1.94	NS

Hosmath et al., (2004a)

character induced by *Bt* possibly leads to a reduced water requirement for *Bt* hybrid and the reduction may be equal to saving of one irrigation. Water requirement of *Bt* cotton at Coimbatore under drip irrigation was 764.8 mm, while in furrow irrigation was 917.2 mm, resulted a water saving of 16.6per cent. Whereas the water requirement of Non *Bt* cotton under drip irrigation was 789.8 mm (6.75per cent) compared to furrow irrigation (967.2 mm) with a water saving of 18.3per cent.The Water Use Efficiency (WUE) in *Bt* Cotton and Non *Bt* Cotton under drip irrigation with fertigation with water soluble fertilizer (WSF) at 100per cent RDF was 4.83 kg /ha mm (64.8per cent increase) and 3.76 kg /ha mm (57.9per cent increase) respectively over furrow irrigation (Asokaraja et al .,2011). Drip fertigation with WSF (drip at 125per cent WRc + fertigation at 100per cent RDF) has recorded higher yield of 39.6 q /ha (47.34per cent higher over control) in *Bt* cotton. Drip fertigation with conventional fertilizer (CF) has recorded 33.1 q /ha (23.24per cent increase over control). In case of Non *Bt* Cotton, the seed cotton yield under drip fertigation with WSF (100per cent dose) was 31.04 q /ha, which was 42.19per cent higher over control and 20.79per cent higher over fertigation with CF (Gokila *et al.,*2011). The yield of *Bt* cotton under drip fertigation with water soluble fertilizer (WSF) at 100per cent dose was 39.6 q /ha which is 27.64per cent increase over non *Bt* cotton (Muthukrishnan *et al.,* 2011). *Bt* cotton sown under irrigated condition (irrigation applied at three critical growth stages of cotton) significantly improved the seed cotton yield (33.71 q/ha) over rainfed condition (15.00 q/ ha) (Dadgale et al 2014) . The studies proved that *Bt* cotton is more efficient in term of water used.

Weed management : *Bt* technology may favor development of super weed by gene transfer to wild relative species. Out crossing of transgenic to wild or weedy crop relatives has become one of the most debated environmental concerns related to transgenic plants. In India, cotton has only one close weed relative, *Gossypium stocksii*, found at northern Gujarat where cotton is not cultivated (Manjunath, 2007

). Pollen dispersal from transgenic cotton is also low. Further, the cotton pollen is heavy and cannot move beyond a few meters away from cotton fields. Therefore, currently the possibility of gene transfer and the development of super weed is a remote possibility. Even in other countries and with other *Bt* crops there is no evidence that super weeds have even developed over the past decade (Manjunath,2007). Pendimethalin @ 1.0 kg /ha followed by a hoeing at Faridkot (Punjab), pendimethalin or prometryn @ 1.5 kg/ha at Sriganganagar (Rajasthan), pendimethalin or trifluralin @ 1.5 kg/ha as pre-plant along with one hand weeding (HW) at 35 DAS for controlling *Trianthema* spp at Sriganganagar (Rajasthan), Galaxy 45 EC @ 2 l/ ha (a ready mix of clomazone @15 per cent w/w + pendimethalin @30 per cent w/w) along with one HW and two intercultural operations with *dora* / *kulpa* at Indore (M.P.) and hand weeding alone at Khandwa (M.P.) and Rahuri (Maharastra) were optimum for higher yield and net return (AICCIP, 2004).

In south, fluchloralin as pre-plant incorporation @ 1 kg/ha along with an interculture and HW at 25 DAS or two HW at 25 and 50 DAS under Lam, Guntur condition, HW or diuron @ 1-1.25 kg/ha along with HW at 30 DAS at Dharward, and two HW at 25 and 50 DAS or pendimthalin or fluchloralin @ 1.5 kg/ha along with two intercultural operations at 30 and 45 DAS at Raichur (KTK) are optimum. At Coimbatore (T.N.), HW twice at 20 and 40 DAS and galaxy @ 2 l/ha or fluchloralin @ 1 kg/ha as pre-emergence are effective (AICCIP, 2004).Pendimethalin @ 1.0 kg /ha as pre-emergence + two hand weeding at 30 and 60 DAS is effective, efficient and economical to control weeds in *Bt* cotton (Usdadia *et al.,* 20011). The highest weed control efficiency and BCR (2.37) were recorded by using the mixture of glyphosate 1kg /ha and pendimethalin 1kg /ha under Stale Seed Bed Technique followed by one hand weeding at 35 to 40 days after sowing in *Bt* cotton (Narayana *et al.,* 2011)

Cropping system

Inter cropping : Cotton is widely spaced, and slow growing in the initial stages for a relatively longer duration offers a vast scope for raising suitable intercrops. When compared to conventional cotton, *Bt* cotton can alter the pest population of cotton ecosystem.This situation warrants the retesting of recommended intercrops of cotton to find out their suitability. *Bt* cotton and *bhendi* intercropping registered the maximum seed cotton equivalent yield (4450 kg/ha) and land equivalent ratio (2.76) (Sankaranarayanan *et al.,* 2004). Relay cropping of *Bt* cotton with *rabi* sorghum, bengal gram, safflower, sunflower, linseed, lentil, peas could not produce any economic yields. But relay with hybrid castor followed by sunflower planted at 45 x 22.5 cm during fag end of September is successful. *Bt* cotton based innovative intercropping systems were evaluated at Nagpur and found that feasibility and profitability of intercropping of green leafy vegetables, cow pea, radish and cluster bean with *Bt* cotton recorded the mean seed cotton yields were 16,19,18,and 17.3 qt /ha, net profits of Rs. 20,000, 37,000, 42,000, 43,000 and benefit cost ratio of 1.97, 2.95, 4.43 and 5.57 respectively. Angrej Singh and Thakar Singh (2015) reported that maximum seed cotton equivalent yield recorded under *Bt* cotton + fodder maize intercropping system (2.61 t/ha) was 10.6, 17.6, 27.3 and 39.6per cent higher than *Bt* cotton + fodder cowpea, *Bt* cotton + summer mungbean, *Bt* cotton + long melon and *Bt* cotton + fodder pearlmillet, respectively. Raman Jeet Singh, and Ahlawat(2014)

suggested that inclusion of legume and organic manure in transgenic *Bt*-cotton–wheat system is a sustainable practice for combating escalating prices of N fertilizers with environmental issues and instability of transgenic hybrids in south Asian countries.

Multi tier cropping : *Bt* cotton based multi tier vegetables intercropping system was developed at CICR, Coimbatore. In three tier system the highest seed cotton equivalent yield (43.5 q/ha) was registered with multi-tier system of cotton + radish+ amaranthus, where intercrops were planted between cotton rows, which was 99per cent higher than that in sole cotton (21.9 q/ha) and also registered the highest gross return (Rs.84,908/ha.) and net return (Rs.55,832/ha.) and benefit cost ratio (2.9) (Sankaranarayanan *et al.,*2008). Multi tier (three intercrops) vegetables intercropping of cotton +radish + cluster bean+ beet root system produced as much as seed cotton yield (25.45 q / ha) as compared to sole cotton (26.15 q /ha) in addition to vegetable yield of radish (6660 kg / ha) , cluster bean (4536 (kg /ha) and beet root (5671 (kg /ha) . Multi tier system was calculated the highest relative production and economic efficiency and land equivalent ratio of 182.2per cent, 308.7 per cent and 2.2 respectively as compared to sole cotton.

Refuge crop : Cultivation of refuge crop is mandatory for cultivation of *Bt* cotton.Insect resistance management is very important to conserve the *Bt* technology for long term benefits and therefore, refuge crop system is advocated. Non *Bt* cotton refuge is practiced in other countries like USA, and Australia and have also been recommended by Govt. of India. The refuge crop could be 20 per cent of the *Bt* cotton with the intervention of plant protection measures, when required or 5 per cent of the area without providing any chemical protection. As a partial modification of this regulatory requirement, the GEAC has now permitted planting strip crop of pigeonpea as a refugia. Thus strip cropping of cotton + pigeonpea, a widely adopted system in Central India (Blaise *et al.,* 2005) will continue even with *Bt* hybrids. Isbell (2000) reported that net loss in return due to the 30 per cent refuge is US$ 29.8/ha. In Argentina alternate host crops, such as corn, soybean and sorghum, which provide an additional non *Bt* refuge (ICAC,2007). The Environment protection Agency (EPA) of USA has approved a natural refuge for Bollgard II, if planted in the states east of Texas. However, natural refuge is not allowed wherever pink boll worm *(Pectinophora gossypiella)* is a major pest, because alternate host crops and plants do not produce a large enough susceptible population (ICAC, 2007). Planting of *Bt* cotton in 80 per cent of area and allotting of remaining 20 per cent area to marigold/okra-chickpea is recommended as alternate strategy for refuge (non *Bt* cotton) by UAS,Dharwad (Anonymuos,2008a).

Sequential cropping : *Bt* cotton hybrids matured earlier by 15 to 30 days than local checks. This advantage with *Bt* cotton can be exploited for making timely sowing of succeeding rabi crop (e.g. wheat, mustard and maize) in cotton based cropping system of North and Central zone and timely planting of kharif rice crop in rice fallow cotton tract of South Zone. Early maturity induced by *Bt* technique, restricting the completion of *Bt* cotton harvest before December, which favored for double cropping in certain regions. The cropping system is shifted from monocropping to *Bt* cotton- Maize in Andhra Pradesh and *Bt* cotton- wheat/ Summer ground nut in Maharashtra and

Gujarat states. However, there is no shift is observed in existing Soybean- hybrid cotton + pigeon pea system of Central India. Nalayini *et al.,* (2009) found that poly-mulched cotton-maize system recorded the highest net return (Rs 74,178/ha) and benefit cost ratio (1.68) as against the conventional system (Rs 29,863 and 1.04).In the north zone, even with *Bt* hybrids, cotton-wheat system remained more efficient and profitable than either cotton-barley or cotton-mustard systems (TMC, 2008).

Cotton-wheat : The determinate nature of *Bt* cotton enables timely sowing of wheat leading to greater system productivity of this most widely practiced cropping system (1.5 m ha) of north-west plains zone. Early sowing of wheat (8th Nov.) gave significantly more grain yield (41.22 q/ha) than the late sowing (20th Dec.) (37.58 q/ha) (Shirpurkar *et al.,* 2008). The grain yields of wheat decreased with each day delay in sowing might be because of shorter period available for anthesis and grain development. Reduction in wheat yield due to late sowing has also been reported by Sardana *et al.,* (2002).The adoption of *Bt* technology increased the average productivity of cotton from 300 to 558 kg lint/ha in the cotton-wheat production zone. Gangaiah, and . Ahlawat (2014) reported that two *Bt* cotton hybrids did not markedly alter the performance of succeeding wheat. However,the system productivity (cotton-equivalent yield, CEY) of *Bt* cotton–wheat was 24.2per cent (4.72 tonnes/ha) higher than the non-*Bt* cotton–wheat system (3.80 tonnes/ha).

Fibre quality : Besides yield improvement, quality of cotton fibre is the other important component that has a bearing in any cotton research programme. Jenkins *et al.,* (1991) in the first field test of transgenic cotton lines reported that yield and fibre properties were in the range of the adapted cultivar Similarly, Ethridge and Hequet, 2000 could not find differences in micronaire, uniformity ratio, strength and elongation measured in High Volume Instrument as a result of transgenic technology. The quality parameters viz., 2.5per cent span length, maturity ratio, uniformity ratio, micronaire, fibre elongation and fibre quality Index were not different in hybrids or *Bt* versus non-*Bt* (Sankaranarayanan *et al.,* 2008). None of the physical and quality parameters differed with NCS 145 *Bt* and NCS 145 hybrid

Table 5. Seed cotton yield (kg/ha) and quality parameters in *Bt* and non *Bt* hybrids

Parameters	Yield span (kg/ha)	2.5 per cent length (mm)	Uni formity-ratio	Micro-naire value	Maturity (%)	Strength (g/tex)	Elong-ation (%)
MECH 184 *Bt*	2183	29.13	46.33	3.85	66.93	24.17	5.58
MECH 184 non *Bt*	715	29.3	45.83	3.72	65.48	23.07	5.60
MECH 162 *Bt*	1912	25.16	47.83	3.67	66.17	19.18	5.87
MECH 162non *Bt*	1077	24.50	48.50	3.82	66.43	18.75	5.83
MECH 12 *Bt*	1935	27.53	48.17	3.53	65.07	23.95	5.13
MECH 12 non *Bt*	634	27.95	48.17	3.68	65.77	22.97	5.67
SEM +	193.5	0.41	0.41	0.09	0.80	0.60	0.11
CD at (p=0.05)	647	1.38	1.36	NS	NS	20.2	0.34

Hallikeri *et al.,* (2004)

under rainfed conditions in vertisols at LAM, Guntur (Bharathi *et al.,* 2011).Performance trial conducted at Dharwad (Table 6) under protective irrigated condition (Hallikeri *et al.,* 2004) revealed that quality parameters were not influenced by *Bt* gene. Ginning percentage and halo length, *Bt* cotton hybrids expressed better performance which was on par with their respective non-*Bt* hybrids (Ansingkar *et al.,* 2005). There was no difference among micronaire, 2.5per cent span length, immature fibre content, neps/g and maturity ratio at the final stage of fibre development. However, the tenacity encountered some change in the final stage of fibre development (Yadav et al, 2012).

Contrary to the general trend, Mayee *et al.,* (2004) reported that *Bt* hybrids exhibited higher ginning percentage over their non *Bt* counterparts. For lint index, greater differences were observed between *Bt* and non *Bt* hybrids. *Bt* hybrids exhibited slightly lesser 2.5 per cent length than their non -*Bt* hybrids. This is possibly due to the shortening in the crop duration. However, these differences noted are not of much consequence as they were in the acceptable range. In general, the assessment of fibre quality parameters of *Bt* cotton hybrid indicate that the fibre properties of *Bt* cotton hybrids were in the range of the adapted cultivar with few exceptions.

CONCLUSION

Blaise et al (2014) reported *Bt* cotton that besides providing resistance against lepidopteron pests, the *Bt*. hybrids matured earlier, were more determinate, had a rapid leaf area development, retained more early set fruiting forms. Some prominent changes accompanying this technology have been, compulsory seed treatment with imidachloprid, advancement in sowing wherever supplemental irrigation was available, a delay in sowing in the traditional rainfed till the soil profile absorbs sufficient moisture, application of atleast 25per cent higher fertilizer dose, shift towards balanced fertilization, foliar application of K and micronutrient mixtures. Adoption of drip irrigation and fertigation, more widespread use of booth pre-emergence (pendimethalin) and post-emergence herbicides (quizalofop ethyl and pyrithiobac sodium) practice of stale seed bed technique for weed management, pruning and extension of crop duration are some other agro techniques which are being adopted by *Bt* cotton farmers.

The steep increase in adoption of *Bt* hybrids across the country by numerous farmers have proved that the technology is well accepted by Indian farmers. Around 95per cent of the cotton area is now under *Bt* hybrids and this acreage has perhaps saturated. Over the last few years despite increase in area under *Bt* hybrids the productivity at national level has not improved. The morphological and physiological changes in these introgressed *Bt* cultivars offer an excellent opportunity for agronomic manipulation. Hence, further improvement in the productivity of cotton and cotton based production systems can only be expected by fine tuning its agronomic practices. Several opportunities for enhancing yields with *Bt* cotton have been documented. Adoption of these technologies may help to improve *Bt* cotton yields.

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Status of cotton harvesting machinery in India

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Abstract : In India, harvesting of cotton is done manually and cost of cotton harvested by hand is quite high and increasing further each year. Hence, there is an urgent need to develop a suitable cotton harvester for small and marginal farmers in India. The study depicts the status of the cotton harvesting machinery in India and covers the experimental research carried out by scientists for development and evaluation of cotton harvesters of different picking type such as pneumatic/suction picking and mechanical picking. Pneumatic knapsack cotton picker were tested by different scientists and found that the maximum picking efficiency *i.e.* 97.5 per cent with trash content 14 per cent with single nozzle while the picking efficiency with double picking nozzle was 95 per cent with 8.80 kg/hr output capacity. The mechanical picking was done by scientists with locally developed and commercially available cotton harvesting machines. The average values of forward speed, effective field capacity, fuel consumption, total harvesting loss, mechanical picking efficiency and picker efficiency of spindle type cotton picker were 2.20–3.38 km/h, 0.278–0.563 ha/h, 22.0–24.0 l/h, and 14.29–31.74, 55.6–83.1 and 68.3–85.7 per cent, respectively. The average picking efficiency and picking capacity of developed self propelled cotton stripper was observed in the range of 76-80 per cent and 135-325 kg/hr, respectively. The observed value of seed-cotton was observed in the range of 74-80 per cent by using boll crusher/seed-cotton extractor.

Cotton is cultivated in tropical and subtropical countries, namely China, USA, India, Pakistan, Uzbekistan, Turkey, Brazil, Greece and Egypt. These countries with temperatures ranging between 11°C and 40°C contribute about 80 per cent of the global cotton production (Anonymous, 2010). Major crop production operations for cotton include field preparation, planting, weed control, spraying and picking. Amongst all cotton picking is the most difficult, tiresome and tedious operation. The labour requirement for cotton picking reported to be about 500 man h/ha. It was not only tedious but also ten times costlier than irrigation and about twice more costlier than the weeding operation (Prasad and Majumdar, 1999). A grown up person can pick about 15-20 kg/day of seed cotton, compared to an average pick of 870-2180 kg/day by a single row spindle type picker (Sandhar, 1999). Cotton is mostly picked manually in most of the developing countries. It is a tedious and labour intensive task mostly performed by ladies. In advanced countries like USA, Australia, Brazil and Russia, cotton picking is carried out mechanically by cotton pickers (the most commonly used machines) or cotton strippers. In India too, harvesting of cotton is done manually and cost of cotton harvested by hand is quite high and increasing further each year. Hence, there is an urgent need to develop a suitable cotton harvester for small and marginal farmers in India. The study depicts the status of the cotton harvesting machinery in India and covers the experimental research work carried out for development and evaluation of cotton harvesters of different type of picking such as pneumatic/suction picking and mechanical picking.

Pneumatic picking type cotton harvester : Sandhar and Goyal, (2003) tested suction principle for cotton picking. The cotton picker consisted of a blower, tank and suction hose. The components were mounted on a frame hitched to the three point linkage of a tractor. The drive to the blower was provided from tractor PTO through gear box. The picker had the picking efficiency in the range of 63.4 to 77.5 per cent at suction pressure of 240 mm of water head at the blower speed of 2875 rpm. The machine picked the fully open bolls but left the cotton bolls which were either infected or rigidly adhered to the carpel. The infection dependent upon the season but adherence depended upon variety. The manually picked cotton after machine operation ranged from 22.5 to 36.6 per cent.

Rangaswamy *et al.,* (2006) conducted a study for optimization of machine parameter of pneumatic knapsack cotton picker. The dimensions of the machine components viz., pickup diameter, filter type, filter height, capacity of collection drum and speed of aspirator were optimized through statistical analysis. The combination of a 25 mm diameter pickup pipe, a nylon mesh filter 225mm high, a 25 litre collection drum and a 5500 rpm aspirator speed developed maximum pressure. It was found that the field capacity for the first picking (4.93 kg/h) was less than that for the third picking (5.07 kg/h). The picking efficiency was lower (96.35%) in the first picking and higher (97.48%) in third picking. The trash content in the machine picked cotton was a maximum of 13.97 per cent in third picking. The saving labour cost, time and energy in machine picking compared to conventional picking was 9.00, 75.00 and 68.23 per cent, respectively.

Ankit (2008) developed a tractor operated pneumatic cotton picker for cotton picking machine comprises a vacuum pump provided to create a predetermined vacuum and a cotton picking unit having single nozzle. Preliminary testing of the developed picking aid was done in the field using various combinations of the picker end diameters (20, 25, 32 and 40 mm) and suction pressures (25, 30, 35, 40, 45 and 50 mm of Hg) to study their effect on picking efficiency, trash content and output capacity. Maximum picking efficiency of 96.3 per cent was achieved at 25 mm of picker end diameter with suction pressure of 45 mm of Hg. Minimum trash content of 0.65 per cent was observed at 20 mm of picker end diameter with suction pressure of 30 mm of Hg. Maximum output capacity of 6.25 kg/h was achieved at 25 mm of picker end diameter with suction pressure of 45 mm of Hg. Picking aid was then evaluated for long term field trials with optimized picker end diameter (25mm) at varying suction pressures (35, 40, 45 and 50 mm of Hg) for two stages of picking. Though the picking efficiency and output capacity was maximum at 50 mm suction pressure for first picking but trash content increased at this suction pressure. However minimum trash content of 5.7 per cent was obtained at 35 mm of Hg with picking efficiency of 93.9 per cent and output capacity of 4.2 kg/h. Similarly, for second stage of picking, minimum trash content of 4.39 per cent was observed at 35 mm suction pressure with picking efficiency of 92.8 per cent and output capacity of 4.01 kg/h.

Patil *et al.,* (2015) developed a knapsack type cotton plucker to suit for farmers cultivating cotton on small scale consisting of prime mover, blower, filter, pick up pipe and collection drum. A polypropylene container of 50 liter capacity was fixed on the frame to collect cotton. Filter was used inside collection drum to restrict the entry of cotton inside the aspirator. Two lightweight aluminum pipe of 50 mm diameter were used

as the pick up pipe. Total length of suction pipe and pick up pipe was kept as 1580 mm. The performance of developed knapsack type cotton plucker was tested in a laboratory in terms of fuel consumption, picking efficiency, trash content and output capacity for three different type of drum (A, B and C model) and four different speed of blower (4200, 4700, 5200 and 5700 rpm). Results indicate that the fuel consumption ranged from 0.270 to 0.702 l/h, picking efficiency from 91 to 96 per cent; trash content from 2.07 to 8.03 per cent and output capacity from 4.75 to 9.78 kg/h. On the basis of laboratory results B type drum was selected for field evaluation at 5200 rpm speed of blower. The average fuel consumption (l/h), picking efficiency (per cent), trash content (per cent) and output capacity (kg/h) was observed as 0.603, 94.79, 5.77 and 8.84 respectively.

Mechanical picking type cotton harvester : Prasad *et al.,* (2007) evaluated a two row self-propelled cotton picker at different locations in India. The performance of John Deere 9935 cotton picker was evaluated at PAU Ludhiana and CICR, Nagpur. The mean values of forward speed, effective field capacity, total harvesting loss, mechanical picking efficiency and picker efficiency were 2.62 km/h, 0.28 ha/h, 23.62, 75.7 and 76.4 per cent, respectively for the experiments conducted at CICR, Nagpur. The mean values of forward speed, effective field capacity, fuel consumption, total harvesting loss, mechanical picking efficiency and picker efficiency were 2.20–3.38 km/h, 0.278–0.563 ha/h, 22.0–24.0 l/h, and 14.29–31.74 per cent, 55.6–83.1 per cent and 68.3–85.7 per cent, respectively for the experiments conducted at PAU, Ludhiana. The cultural practices and staggered blooming characteristics of present Indian cotton varieties poses challenge to engineers in mechanization of cotton picking. With the advent of new genotypes, it may be possible to introduce mechanical cotton pickers successfully.

Singh *et. al.,* (2014) evaluated commercially available low cost handheld portable cotton pickers having two types of mechanisms *i.e.* chain and roller and their performance were compared with manual picking. Labour required during manual picking is significantly lower than roller type cotton picker at 5 per cent level of significance. There was no significant difference in the picking rate among chain, roller and manual picking at 5 per cent level of significance. The percentage of trash content for both chain and roller type cotton pickers was higher *i.e.* 11.52 and 10.44 per cent as compared to trash content of 7.43 per cent measured for cotton picked manually.

Sharma *et. al.,* (2015) developed and evaluated a self propelled finger type cotton stripper to pick/harvest the local high density and dwarf cotton varieties mechanically. In the developed stripper, stripping fingers of 70 cm length were welded to the front part of engine frame at an angle of 210. The width of the developed head was 64 cm. A rotating paddle/kicker, having a speed in the range of 120-250 rpm, was designed to push the stripped materials (cotton bolls *i.e.* opened and closed along with sticks and burs) in to the collecting tank. A collecting drum/tank, having capacity 15-20 kg, was attached just behind the cotton stripper head for collecting stripped cotton materials. The developed prototype was evaluated on F 2383 and RCH 773 cotton varieties to observe its performance. The average value picking efficiency and picking capacity of developed cotton stripper was observed to be in the range

of 76-80 per cent and 135-325 kg/hr, respectively. The observed value of seed-cotton was observed in the range of 74-80 per cent by using boll crusher/seed cotton extractor, operational at Bathinda in Punjab.

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Agronomy of transgenic and non transgenic cotton in India

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Cotton is an important commercial crop and plays a vital role in Indian economy and is primarily grown for its fibre which is the most important raw material for textile industry.Bt cotton is being extensively cultivated on large area in India. About 60 million people in India are engaged directly or indirectly in textile industry. The Worlds, most important non food agricultural commodity, was one of the first vegetable fibers used for textile purpose Even today ,it is unchallenged as a natural textile fibers , and entering our daily life in a variety of way and is the most valued amongst several hundred fiber yielding plants known to mankind.

Balanced fertilization has been proved to be kingpin in agricultural production under different farming situations and contributed to nearly 50 per cent in overall increase of agricultural production. The soils in the northwestern states are not only thirsty but also hungry. The soils are deficit in nitrogen and phosphorus whereas they are well supplied with potassium in major areas of these states. The deficiencies of available sulphur, boron, zinc and iron are also emerging in the region on large scale. Therefore, it is very essential to use fertilizers in balanced proportion due to risk involved, particularly in rainfed farming. The rainfed crops are deprived of the fertilizers resulting into low crop yields in Madhya Pradesh, Maharashtra and Rajasthan. Even under assured rainfall situation in Maharashtra and Madhya Pradesh, the use of fertilizers is low as compared to national average. Promotional efforts are therefore, very much essential in these areas.

The nitrogen fertilization alone prolongs the crop maturity and invites the problems of pests and diseases, whereas phosphorus and potash application hasten the crop maturity, increase the efficiency of available soil moisture in crop production and help the crops to escape from the stress conditions in the later period of the crop growth. It is therefore, essential to adopt a balanced fertilizer scheduled for higher efficiency and profitability.

The use of mineral fertilizers is the fastest and definite way to improve crop productivity. However, the increases in cost and associated environmental hazards as well as lack of sustainability in yields under application of such fertilizers are constraints in cotton production. Low soil organic matter coupled with deficiencies of nutrition and continuous cotton cropping and management practices are the main reasons for lack of sustainability. This has renewed the interest in the use of organic fertilizers along with inorganic fertilizers. High and sustainability productivity of cotton is associated with balanced nutrition and available nutrient in the soil.

Cropping sequence : A field experiment was conducted at Agriculture Research Station, Sriganganagar revealed that highest net return was recorded with *hirsutum* cotton-wheat cropping systems, followed by *hirsutum* cotton-mustard, but sustainability indices calculated on the basis of

net return was the highest (96.30%) in *arboreum* cotton wheat cropping system. *Hirsutum* cotton mustard gave the highest B:C ratio (1.75) followed by *arboreum* cotton mustard cropping system (1.63) (Nehra & Bhunia, 2002) (Table 1).

A field experiment was conducted at Punjab Agricultural University, Regional Station, Faridkot during 2000-2001 and 2001-2002 seasons the data revealed that cotton-wheat, cotton-barley and cotton-chickpea would be suitable crop rotation to achieve maximum economical yield. Higher water use efficiency was observed red in rotations with pulse crops (Chickpea and fieldpea) followed by rotations with cereal crops (wheat and barley). Therefore, it was concluded that cotton-wheat would be the best crop rotation under adequate irrigation facilities and cotton-chickpea for the less irrigation facilities areas (Singh *et. al.,* 2003) (Table 2).

Table 1. Economics of different cropping systems

Cropping systems	Gross return (Rs/ha)	Net return (Rs/ha)	Sustainability index of net return (%)	B:C ratio
Hirsutum cotton mustard	47341	30111	93.3	1.75
Arboreum cotton mustard	46008	28508	93.0	1.63
Hirsutum cotton wheat	50860	30749	95.2	1.53
Arboreum cotton wheat	49033	28651	96.3	1.40

Table 2. Economics and water use efficiency of different rotations during two crop season at Faridkot.

Rotations	Mean Net returns	WUE kg/ha/ mm
Cotton Fellow	15928	3.82
Cotton Wheat	36534	3.89
Cotton Chickpea	35083	4.44
Cotton Barley	31368	3.77
Cotton Mustard	20011	3.20
Cotton Fieldpea	23265	3.96

Time of sowing: One of the important factors in enhancing the seed cotton yield is the time of sowing. Time of sowing affects plant growth and fruiting through its effects on the microclimate of the crop. Lint yield is a product of number of mature bolls produced/unit area. Late sowing resulted in decrease in opened bolls, increased pest attack and reduction in yield. Early sowing of the crop helped in the timely sowing of the succeeding *rabi* crops. Planting time differs from place to place for obtaining higher yields. In Haryana, Punjab and Rajasthan, sowing is recommended in the month of April to mid May similar results were recorded by Kumar *et al.*, (2014), (Nehra and Chandra 2001) and (Anonymous 2000) (Table 3, 4 and 5).

Spacing and fertilizer: In RCH 134 *Bt*, 108 x 60 cm row spacing gave significantly higher seed cotton yield at Sriganganagar, where as 100 x 45 cm spacing shows its superiority at Hisar but at par results have been observed at Ludhiana and Faridkot. As regard fertilizer levels, 100 per cent RDF seems to be optimum at Faridkot and Sriganganagar, where as 125 per cent RDF gave significantly higher seed cotton yield at Hisar but no significant difference in seed cotton yield was noticed at Ludhiana (Table 6).

Central zone : The spacing of 90x90 cm

Table 3. Performance of different *Bt* cotton (*Gossypium hirsutum* L) hybrids under varying dates of sowing

Treatments	Seed cotton yield (kg/ha)	Bolls/ plant	Boll weight (g)
Dates of sowing			
3rd week of April	3166	50.61	3.60
1st week of May	2905	48.02	3.64
3rd week of May	2642	42.51	3.88
1st week of June	2345	37.69	3.85
C.D. (p=0.05)	389	8.01	NS
Genotype			
RCH-134	2630	37.55	4.14
NCS-855	2963	49.05	3.60
Bio-648	2702	47.53	3.54
C.D. (p=0.05)	228	5.05	0.41

Table 4. Effect of different sowing dates on the yield of American cotton at Sriganganagar

Sowing date	Seed cotton yield (kg/ha)		**Mean**
	1997	1998	
1st May	1995	2166	**2081**
15th May	1653	1710	**1682**
30th May	655	860	**758**
CD (p=0.05)	111	137	**—**

Table 5. Effect of different sowing dates on the yield of American cotton at Abohar

Sowing date	Seed cotton yield (kg/ha)			**Mean**
	1998	1999	2000	
6-8 April	1068	1965	1848	**1627**
20-22 April	1086	2133	2154	**1791**
4-7 May	701	1867	1849	**1482**
18-19 May	624	1677	1319	**1207**
1-8 June	226	751	1135	**704**
CD (p=0.05)	146	219	256	**—**

Table 6. Effect of spacing and fertilizer level on *Bt* cotton hybrid (RCH 134 *Bt*)

Treatments	Seed cotton yield (kg/ha)			
	Faridkot	Ludhiana	Hisar	Sri ganganagar
Row spacing (cm)				
67.5 x 60	-	-	3827	2144
67.5 x 75	2892	2293	-	-
100 x 45	-	-	3838	-
100 x 60	3151	2251	3530	-
100 x 75	3003	2079	-	-
108 x 45	-	-	-	2336
108 x 60	-	-	-	2421
CD (p=0.05)	NS	NS	170	125
Fertilizers levels (kg/ha)				
RDF (75%)	2725	2188	3443	2098
RDF (100%)	3117	2174	3740	2372
RDF (125%)	3204	2260	4011	2430
CD (p=0.05)	254	NS	170	125

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at Rahuri and Khandwa, 90x45 cm at Indore and Akola, 120x45 cm at Junagarh and Surat and 90x60 cm at Nanded was found optimum for RCH-2 Bt hybrid. As regard level of fertilizer,125% RDF at Indore, Nanded, Surat, Akola and Rahuri gave the highest seed cotton yield where as 75 per cent RDF seems to be optimum at Khandwa and Junagargh**.** (Table 7).

South zone : The spacing of 90x45 cm showed its superiority at Lam, Nandyal and Siruguppa where as 90x60 cm was found better at Coimbatore and Srivilliputtur and all the spacing were *at par* at Dharwad in Bunny *Bt.* As regard level of fertilizer 125 per cent RDF gave highest seed cotton yield at all the stations except Siruguppa where 100 per cent RDF seems to be optimum (Table 8).

Fertilizer:

Nitrogen: Cotton crop responds well to the

Table 7. Effect of spacing and fertilizer level on *Bt* cotton hybrid (RCH 2 *Bt*)

Treatments	Seed cotton yield (kg/ha)						
	Khandwa	Indore	Nanded	Junagarh	Surat	Akola	Rahuri
Row spacing (cm)							
90x45	-	1421	2039	-	-	1210	2120
90x60	1789	1357	2380	-	-	1132	2314
90x90	1871	1177	2133	-	-	950	2392
120x30	-	-	-	-	2122	-	-
120x45	-	-	-	3092	2733	-	-
120x60	-	-	-	2594	1589	-	-
120x90	2206	-	-	1679		-	-
CD (p=0.05)	152	47.3	96.4	501	373	90.1	NS
Fertilizers levels (kg/ha)							
RDF (75%)	2207	1235	1966	2398	1808	1033	2160
RDF (100%)	1855	1309	2174	2444	2148	1109	2296
RDF (125%)	1804	1411	2411	2523	2488	1149	2369
CD (p=0.05)	152	43.4	172	NS	196	64.2	119

Table 8. Effect of spacing and fertilizer level on *Bt* cotton hybrid (Bunny *Bt*)

Treatments	Seed cotton yield kg/ha					
	Lam	Nandyal	Siruguppa	Coimbatore	Dharwad	Srivilliputtur
Row spacing (cm)						
90x30	3664	-	-	-	-	-
90x45	3700	1990	2664	2302	2230	2783
90x60	3571	1586	2567	2348	2318	2831
90x90	2912	1192	2267	2115	2349	1962
120x90	-	1241	-	-	-	-
CD (p=0.05)	308	319	65.39	150	NS	365
Fertilizers levels (kg/ha)						
RDF (75%)	3320	1458	2345	2224	2161	2154
RDF (100%)	3409	1452	2605	2217	2324	2569
RDF (125%)	3412	1596	2548	2325	2412	2853
CD (p=0.05)	325	131	105	47.7	94	353

application of fertilizer. Optimum doses of nitrogen, phosphorus and potassium fertilizers required for *arboreum, hirsutum* and *Bt* hybrids are 90 N 20 P_2O_5, 80 N-40 P_2O_5 20 K_2O and 150 N-40 P_2O_5 20 K_2O kg/ha, respectively so as to obtained higher seed cotton yields over other combinations in canal command area of north west Rajasthan. Experiments on nitrogen conducted at ARS, Sriganganagar revealed that for American cotton, 80 kg N/ha was optimum dose, application of N in two equal splits, 50 per cent (40 kg/ha) as basal and remaining 50 per cent at the time of square formation has been found most effective to achieve optimum seed cotton yield. In case of hybrids nitrogen may be applied in three equal splits i.e. 1/3 at basal, 1/

3 at 1st irrigation and remaining 1/3 at square formation stage. In case of phosphorus whole P_2O_5 (40 kg/ha) will be supplied at the time of sowing (Bhardwaj *et al.,* 1999).

Phosphorus: Phosphorus is essential to many processes such as photosynthesis, carbohydrates metabolism and the formation of seed and fibers. Trials (Table 9) involving newly released high yielding varieties responded to P application and 40 kg P_2O_5/ha was adequate for irrigated as well as rainfed cotton Nehra and Yadav (2011).

Potassium: Where the soil is deficient in potassium, application of this nutrient may result in higher yield, improved quality and increased resistance to pest and disease. The application of potasic fertilizer @ 30 kg/ha at sowing resulted in 1.87 q/ha highest seed cotton yield than without potash. Further increase in potash dose could not show its impact on seed cotton yield at Sriganganagar (Table 10). In a long term field experiment conducted at CRS, Sirsa with cotton-wheat rotation it was observed that application of 30 kg K_2O/ha increased the yield of cotton from 13.2 to 14.30 q/ha and thereafter further addition of K_2O did not make any significant improvement as shown in (Table 10).

Table 9. Response of cotton to P application in north zone.

Place	Irrigated/rainfed	Dose giving significant response (kg P_2O_5/ha)	References
Delhi	Irrigated	60	Prasad and Prasad, 1998
Sriganganagar	Irrigated	40	Nehra and Yadav, 2011

Table: 10. Effect of potash application on seed cotton yield

Treatments	Seed cotton yield (q/ha)	
	Srigan-ganagar (Raj.)	Sirsa (Haryana)
Control	19.61	13.20
30 kg K_2O/ha at sowing	21.48	14.30
60 kg K_2O/ha at sowing	21.53	14.52
C.D. (p=0.05)	1.21	1.03

Combined effect of macro and micronutrients on the production of cotton: The combined application of Recommended dose of N, P, K, S and Zn produced higher seed cotton yield over RDF at Faridkot, Sirsa, Sriganganagar,

Table 11. Effect of macro and micro nutrients on yield of cotton under cotton wheat system.

Location	Components (Macro and micro nutrient)	Yield in macro and micro nutrients (kg/ha)	Yield with RDF (kg/ha)	% Increase over RDF
Farid kot	RD-NPKSZn	1995	1816	9.88
Sirsa	RD-NPKSZn	2028	1941	4.48
Kanpur	RD-NPKS	1091	940	16.06
Srigangangar	RD-NPKSZn	1409	1193	18.10

Source: AICCIP Annual Report, 2003-2004

while RD of NPKS produce higher seed cotton yield over RDF at Kanpur (Table 11).

Integrated nutrient management practices on cotton production: Combined application of 10 t FYM/ha along with 50 per cent RDF + foliar spray of nutrients produced significantly higher seed cotton yield at Faridkot,Hisar and Sriganganagar (Table 12).

Table 12. Yield advantage due to integrated nutrient management practices.

Location	Component of INM	Yield in INM (kg/ha)	Yield with RDF (kg/ha)	Grain due to INM (%)
North zone				
Faridkot	RD (50%) + NPK + 10 t FYM/ha + foliar spray	2355	1898	24.08
Hisar	RD (50%) + NPK + 10 t FYM/ha + foliar spray	3596	3391	6.04
Sriganganagar	RD (50%) + NPK + 10 t FYM/ha + foliar spray	3017	2694	11.99

Source: AICCIP Annual report 2003-2004, CICR-Regional Station Coimbatore.

Foliar application of KNO_3 to increase the yield and yield attributes of the cotton : Three sprays of 3 per cent KNO_3 gave the highest seed cotton yield at Akola and Junagarh where as Four sprays of 3% KNO_3 seems to be better at Nanded but at Ludhiana no significant result was observed (Table 13).

Weed control : The treatment weed free check gave significantly highest seed cotton yield at all the locations except Junagarh, Indore and Nandyal. Among the chemical weed control treatments Pyrithiobac Sodium @ 62.5g a.i/ha + Quizalofopethyl 50g a.i/ha 20-30 DAS or 2-4 weed leaf stage +one hoeing at Faridkot, Nanded ,Banswara, Coimbatore and Raichur. Glyphosate @ 1.0kg a.i/ha as directed spray at 45 DAS at Bathinda. Pendimethalin1.0kg a.i/ha+ Quizalofopethyl 50g a.i/ha + one hoeing at Sirsa, Surat ,Rahuri, Bhawanipatna, Srivilliputtar,

Table 13. Effect of foliar application of KNO_3 to increase the yield of cotton at various AICCIP locations of northern and central zone

Treatment	Seed cotton yield (kg/ha)			
	Ludhiana	Nanded	Akola	Junagarh
Control	1088	1280	1353	2235
Two sprays of KNO_3 (2%)	1237	1601	1370	2650
Three sprays of KNO_3 (2%)	1406	1618	1433	3210
Four sprays of KNO_3 (2%)	1476	1952	1488	2646
Two sprays of KNO_3 (3%)	1482	1803	1445	3013
Three sprays of KNO_3 (3%)	1346	1610	1582	3269
Four sprays of KNO_3 (3%)	1324	1766	1435	3029
MOP in four splits	1389	1614	1416	3217
Full dose of MOP at sowing	1490	1757	1401	2469
CD (p=0.05)	NS	201	101	684

Source: AICCIP Annual Report

Nandyal and Lam and Pendimethalin extra @ 0.75 to 1.0 kg a.i/ha as pre emergence or PPI + one hoeing at Sriganganagar shows its superiority over the rest (Table 14 to 16)

Table 14. Seed cotton yield (kg/ha) affected by different weed control treatments in north zone

Selected Treatments	Seed cotton yield (kg/ha)			
	Faridkot	Bathinda	Sirsa	SGNR
T_1 : Pendimethalin @ 0.75 to 1.0 kg a.i/ha as Pre emr or PPI + one hoeing	3208	2129	2415	1800
T_2 : Pyrithiobac Sodium @ 62.5g a.i/ha + Quizalofopethyl 50g a.i/ha 20-30 DAS or 2-4 weed leaf stage +one hoeing	3522	2571	1765	1636
T_3 : Glyphosate @ 1.0kg a.i/ha as directed spray at 45 DAS	2684 2915	2066	1584	
T_4 : Pendimethalin extra@ 1.00 kg a.i/ha as PPI + one hoeing	—-	—	—	2068
T_5 : Weed Free check	3552	2771	3292	2150
T_6 : Weedy check	1918	1850	1384	1111
CD (p=0.05)	435	308	215	260

Source: AICCIP report 2014-2015

Table 15. Seed cotton yield (kg/ha) affected by different weed control treatments in central zone

Selected Treatments	Seed cotton yield (kg/ha)				
	Nanded	Bhawanipatna	Surat	Rahuri	Banswara
T_1: Pendimethalin1@ 1.0kg a.i/ha+ Quizalofopethyl 50g a.i/ha + one hoeing	1393	2003	1261	1844	3525
T_2 : Pyrithiobac Sodium @ 62.5g a.i/ha + Quizalofopethyl 50g a.i/ha 20-30 DAS or 2-4 weed leaf stage +one hoeing	1545	1740	739	1691	3989
T_3 : Glyphosate @ 1.0kg a.i/ha as directed spray at 45 DAS	1882 1505	573	1179	2233	
T_4: Weed Free check	1646	2037	2070	2360	4630
T_5: Weedy check	550	926	443	966	1304
CD (p=0.05)	263.0	372	185	336	283

Source: AICCIP report 2014-15

Irrigation : Six pos20t sowing irrigations are required for American cotton to obtain the optimum seed cotton yield. However, there was no significant different in seed cotton yield between five and six irrigations. The highest seed cotton yield was obtained when first irrigation was applied at 30-35 days after sowing. First week of October was found most suitable for last irrigation in north-west part of Rajasthan.

Irrigation methods: Different methods of irrigation which of common use in cotton crop are flooding, furrow, sprinkler and drip irrigations. Flood irrigation system is being commonly used in the State. However, the studies have shown that irrigation applied in

Table 16. Seed cotton yield (kg/ha) affected by different weed control treatments in south zone

Selected Treatments	Seed cotton yield (kg/ha)				
	Coimbatore	Raichur	Srivilliputtur	Nandyal	LAM
T_1: Pendimethalin1 kg a.i/ha+Quizalofopethyl 50g a.i/ha + one hoeing	1735	1943	2551	4125	3837
T_2: Pyrithiobac Sodium @ 62.5g a.i/ha 20-30 DAS + one hoeing	2082	2167	2373	3603	3784
T_3: Pyrithiobac Sodium @ 62.5g a.i/ha + Quizalofopethyl 50g a.i/ha at 20-30 DAS or 2-4 weed leaf stage +one hoeing	2199	2259	2515	3974	3668
T_4: Weed Free check	2289	2491	2602	3773	4411
T_5: Weedy check	1060	1369	726	2236	2266
CD (p=0.05)	274	428	171	498	529

Source: AICCIP report 2014-15

furrows rather than flooding resulted in 24 per cent saving of water in north-west part of Rajasthan (Anonymous, 1988). Different drip irrigation schedules were tried and results indicated that 0.6ET at Lam, 0.8ET at Faridkot and 1.0ET at Rahuri and Dharward gave significantly highest seed cotton yield where as all the irrigation schedules were found non significant at Indore. As regards fertilizer levels 100 per cent RDN & K gave significantly highest seed cotton yield at Faridkot, Lam and Dharwad whereas 50 per cent RDN and K performed better at Rahuri and Indore respectively (Table 17).

Table 17. Improving use efficiency of inputs (water and nutrient)

Selected Treatments	Seed cotton yield (kg/ha)				
	Faridkot	Rahuri	Lam	Dharwad	Indore
Irrigation regime					
1.0 ET	2172	2646	3889	2952	1934
0.8 ET	2243	2151	4055	2696	1996
0.6 ET	1913	1657	4523	2593	1936
C.D. (p=0.05)	241	409	197	99	NS
Nutrient Levels					
RDN (100%) & K	2295	2163	4226	3082	1990
RDN (75%) & K	2138	2087	3989	2650	1987
RDN (50%) & K	1895	2084	-	2510	1890
CD (p=0.05)	204	NS	197	99	NS

Source AICCIP report 2014-15

Moisture conservation techniques of ET based Drip irrigation in Bt cotton : Moisture conservation techniques of ET based Drip irrigation in Bt cotton trial shows that 0.8ETc drip +poly mulch gave significantly highest seed cotton yield at all the location of South Zone except Lam where 0.6ETc drip +poly mulch out yielded over the rest (Table 18)

Table 18. Moisture conservation techniques of ET based drip irrigation in *Bt* cotton

Treatments	Seed cotton yield (kg/ha)				
	LAM	Indore	Junagarh	Akola	Banswara
Control	3467	926	2131	1411	2048
Polymulching (P)	3729	1199	2116	1616	3167
0.4 ETc drip	4235	1176	2249	1730	3066
0.4 ETc drip + poly mulch	4449	1243	2337	2146	3591
0.6 ETc drip	4403	1250	2205	1893	3456
0.6 ETc drip + poly mulch	4660	1400	2631	2210	3823
0.8 ETc drip	4093	1321	1999	1443	3704
0.8ETc drip + poly mulch	3753	1411	2939	2722	4032
CD (p=0.05)	176	255	370	448	115

Source: AICCIP report 2014-2015

Constraints for low production

- Adverse weather conditions
- Delayed sowing of cotton
- Non-availability of quality seed
- Cultivation of undiscriptive varieties/ hybrids
- Increase incidence of pests and disease
- Indiscriminate use of insecticides
- Spray of blended pesticides
- Role of pesticide dealers

Strategies for improving the productivity :

- Recommended varieties/hybrids should be planted.
- Cultivation of in descriptive varieties/ hybrids should be checked/ banned.
- State Seed Corporation should gear up the production of certified seed in adequate quantity, specially disease and pest resistant hybrids/ varieties.
- The sowing should be completed by 20th May and the State Govt. should ensure canal water supply during the sowing period in north zone.
- Recommended seed rate should be used to maintain optimum plant population in the field to obtained higher yield.
- Balanced use of fertilizer should be used on the basis of soil test. At present N is being used in over dose and P in under dose.
- Excessive irrigation should be avoided.
- Use of NAA @ of 10 ppm be popularize to reduce physiological shedding of reproductive bodies.
- Monitoring and surveillance at department and university level should be strengthened.
- The Chemicals should always be used at their optimum/ recommended doses.
- Avoided mixing different insecticides.
- One row of Bajra/ Maize sould be planted around cotton fields to attracts birds for predation of insect-pests.
- Use of bio-control agents like NPV, Neem products and *Trichogramma* should be encouraged.
- Avoid growing American cotton in citrus orchards and adjoining bhindi crops.
- Uproot and destroy the infected plants upto initiation of fruiting phase.
- Protect the crop against whitefly at 4-5 leaf stage by using recommended insecticide.
- Follow clean cultivation and destroy Kanghi

buti (*Sida* sp) and peeli buti (*Abutilon* sp) which act as collateral host.

- Destroy ratoon cotton plants during the off season.
- Desi cotton varieties are resistant to leaf curl virus. Encourage *desi* cotton in CLCV affected area.

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Characterization of a bioremediation bacterium from cotton fields of Gunupur area, Odisha

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ABSTRACT : *Carbofuran* (2, 2-Dimethyl-2, 3-dihydro-1-benzofuran-7-yl methylcarbamate) is used as a broad spectrum pesticide (which is a systemic insecticide) to control the pests of many crops (like rice, *cotton,* maize, potatoes, pumpkins, sunflowers, pine seedlings and spinach etc,). The compoud gets degraded naturally in the environment. However, if the chemical persists in the soil, it is biomagnified and can cause health hazardous to plants and animals (*Carbofuran* is highly toxic to vertebrates with an oral lethal dose of 8–14 mg/kg in rats and 19 mg/kg in dogs, which were found to be a neurotoxic) including human populations.

We have undergone an experimental study on *Cabofuran* subjected cotton field of GUNUPUR 521175, Odisha by isolating a microbial strain from same cotton field which uses this insecticide as a source of Carbon and Nitrogen for growth. The characterization of the isolated microbe i.e., both the morphological and biochemical tests revealed that the bacterium found to be as *Bacillus brevis.* This bacterium degrades *Carbofuran* to Carbofuran phenol, which is relatively less toxic substance. In growth kinetic experimental analysis the degradation of *Cabofuran* by bacterium after 5 days (grown) culture was found to be 65.08 per cent. It is also suggested by "plasmid isolation experimental studies" that the basic mechanism of degradation capability of this bacterium is due to presence of a specific gene that is present mainly as extrachromosomal [(resistant gene in plasmid which helps in growth on the pesticide's carbon and nitrogen source].

A further experimental study, especially on the molecular biological characterization of the plasmid may help in developing certain specific strains of bacteria for the bioremediation of *Carbomate* group of pesticides *per se.*

A pesticide is a chemical or biological agent that deters, incapacitates, kills, or otherwise discourages pests. They are used in agriculture to control weeds, insect infestation and diseases (Beirne, 1975). Use of pesticide is tremendously increasing day by day in agriculture with an aim of more production. Carbofurans is a broad spectrum pesticide marketed under the trade name "Furadan", is used to control insects in a wide variety of crops including rice, cotton, potatoes etc. Its IUPAC name is 2, 3, dihydro-2, 2-dimethyl, 7-benzofuranyl methyl carbamate, molecular formula**:** $C_{12}H_{15}NO_3$ and molecular mass is 221.25.

It has been reported that over 98 per cent of sprayed insecticides and 95 per cent of herbicides reach a destination other than their target biotic species, including non-target abiotic species that are air, water and soil (Howard, 1996). Pesticides may cause acute and delayed health effects in people who are exposed. Pesticide use raises a number of environmental concerns. Thus, *in situ* removal, modification or detoxification pesticide is important for restoration of the environment. Bioremediation is a technique that involves the use of organisms to remove or neutralize pollutants from a contaminated site (Luck, 1981; Van der Hoek *et al*, 1998). The process uses naturally occurring

organisms to break down hazardous substances into less toxic or non toxic substances. Microorganisms are used to perform such kind of the function of 'bioremediation' are known as 'bioremediators' (Price, 1974). Many strains of microorganisms have been reported for bioremediation may degrade, transform or accumulate a huge range of compounds including hydrocarbons, poly chlorinated biphenyls (PCBS), poly aromatic hydrocarbons (PAHS), pharmaceutical substances, metals etc. (Flaherty and Wilson,1999). The present work is aimed at "isolation and characterization of carbofuran degrading microorganisms" that are isolated from soil. We have also addressed the rate of biodegradation by the isolated microorganism and attempt has also been made for the identification of bacterium gene responsible for the degradation of the carbofuran.

MATERIALS AND METHODS

1. Collection of soil *:* Ten soil samples were collected from B horizon layer from different cotton field located near Gunupur area, Odisha with sterile plastic bags and then brought to the Laboratory for analysis. The soil samples are dried under room temperature to remove excess amount of water present on the samples.

2. Isolation of the bacteria : The isolation of the carbofuran degrading bacteria at the laboratory was carried out by following serial dilution standard procedures (Aneja, 2002). There are three different culture media are prepared for isolation of the bacteria such as minimal medium (MM), Nitrogen free medium (NFM) and Nitrogen free medium with glucose (NFMG) (Cock, 1985).

3. Maintenance of bacteria at laboratory : It is necessary to maintain the bacterium culture at the laboratory for future purpose. Pure culture of the bacteria was maintained at the laboratory using the enriched medium showing maximum growth. The pure culture of bacterium was maintained by following standard slant culture method with little modifications (Singleton, 1999).

4. Characterization of microorganism : The isolated bacterium was characterized according to Bergey's Manual of Determinative Bacteriology (Holt *et al.*, 1994) with minor modifications. Further, the confirmation was with molecular test and the phylogenic analysis of the species was also made.

5. Determination of Growth curve of the bacterium : The pure culture of isolated bacterium is utilized for determination of growth curve. Optical density (OD) of the culture is recorded with the help of spectrophotometer at 600 nm at an interval of 30 minutes. The medium without bacterial inoculation was maintained as control.

6. Isolation of plasmid from the bacterium : Isolation and recovery of plasmid from the fresh culture of bacterium was carried out with alkaline lyses solutions (Sambrook *et al*, 2006).

7. Bioremediation experiments : The isolated bacterium and the carbofuran (TATA FURAN) which is commercially available are inoculated together in a conical flask and then kept in a rotary shaker. The rotary shaker is allowed for continuous agitation for ten days. The conversation of carbofuran to carbofuran-phenyl a less toxic substance are measured with the help of Thin layer chromatography (TLC) and

Gas chromatography and mass spectroscopy (GCMS) at the regular interval of inoculation. The suspension present in the mixed culture were analysed by Gas chromatography and Mass spectroscopy (GCMS). GC"MS (MSGC" 11) instrument with Capillary column of HP"3 (50 mm × 0.521mm, film thickness 0.25Am). 1 ml of extract was carefully injected into GC"MS for analysis. The chemical compositions were identified by comparing their retention indices (RI) and mass fragmentation pattern. The results obtained were recorded. The maximum days of inoculation (DOI) has been chosen after the constant value was obtained.

RESULTS AND DISCUSSION

The collected soil samples showed bacterial colonies in the culture medium at the land to laboratory experimentation. All the culture media used have developed the bacterial colonies. Table 1 represented the appearance of bacterial colonies in the different kinds of culture media. The numbers of colonies were ranged between 35 and 80 after 5 days of growth bacterial culture medium as specified (Table 1). The number of bacterial colonies were more in designed culture medium containing free of nitrogen source but with glucose as carbon source (80 in number), followed by designed culture medium with free of carbon containing 53 colonies in number. From the both the growth sources of the experiments on developments of the bacterium etc., are identified this microbe as *Bacillus sp*. Growth curve of isolated bacterium strain is established in the batch culture with the Hi-media nutrient broth (Fig. 1). As shown in Fig. 1 the bacterium showed regular sigmoid growth curve with lag phase up to 90 minutes, followed by log or exponential phase up to 250 minutes. After exponential phase the bacterium reached to stationary phase. The plasmid was isolated from the exponentially grown (mid log) culture and isolated

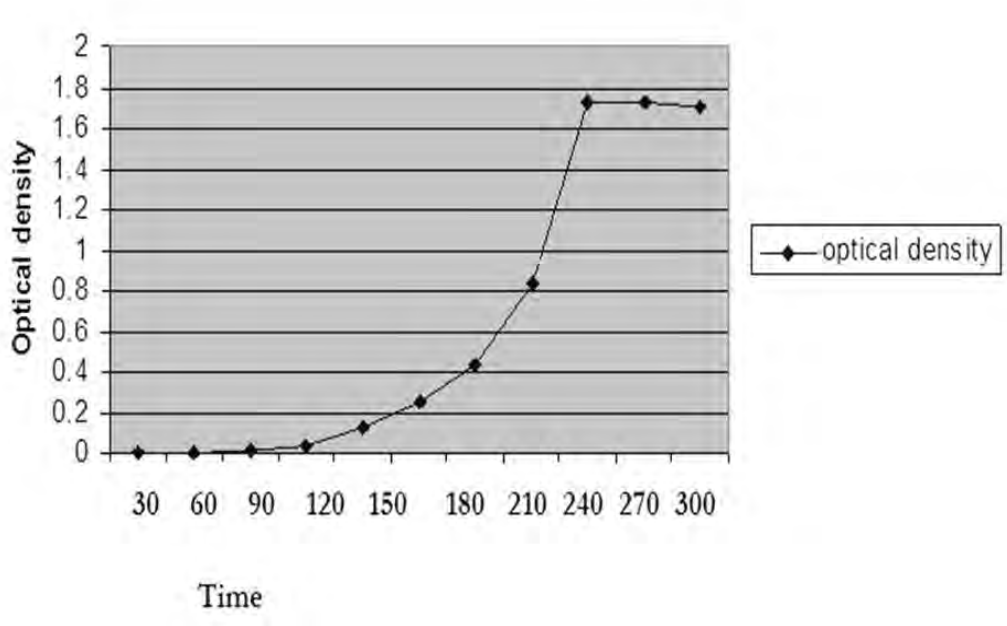

Fig. 1. The growth curve of the isolated bacterium in the batch culture

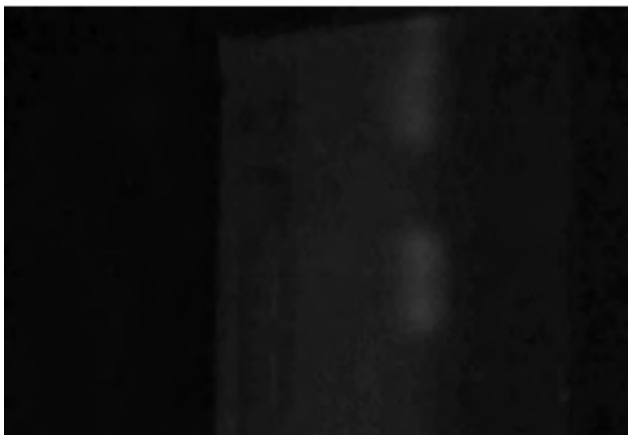

Fig. 2. Appearance of single band of plasmid on agarose gel electrophoresis

plasmid was detected through horizontal electrophoresis. A prominent single band was observed upon staining with ethidium bromide (Fig. 2). This indicated that the bacterium had a single or the different plasmids of same molecular weight. In the studies of carbofuran degradation, where the bacteria were grown in presence of pesticide and it was recorded that with in 7 days, in mixed culture, there the convertion of carbofuran to its metabolic inactivated product was observed (Table 2). The initiation of degradation of the compound was observed at (24 hrs of culture) stationary phase,

Table 1. Appearance of bacterial colonies in the culture medium. Data represent the mean of three replicates.

Days after culture	Designed culture medium with free of nitrogen	Designed culture medium with free of nitrogen with glucose	Designed culture medium with free of carbon
1	00	00	00
2	10 ± 2	18 ±1	14 ± 2
3	25 ± 2	29 ± 3	26 ± 2
4	35 ± 1	42 ± 2	39 ± 1
5	35 ± 1	80 ± 1	53 ±1

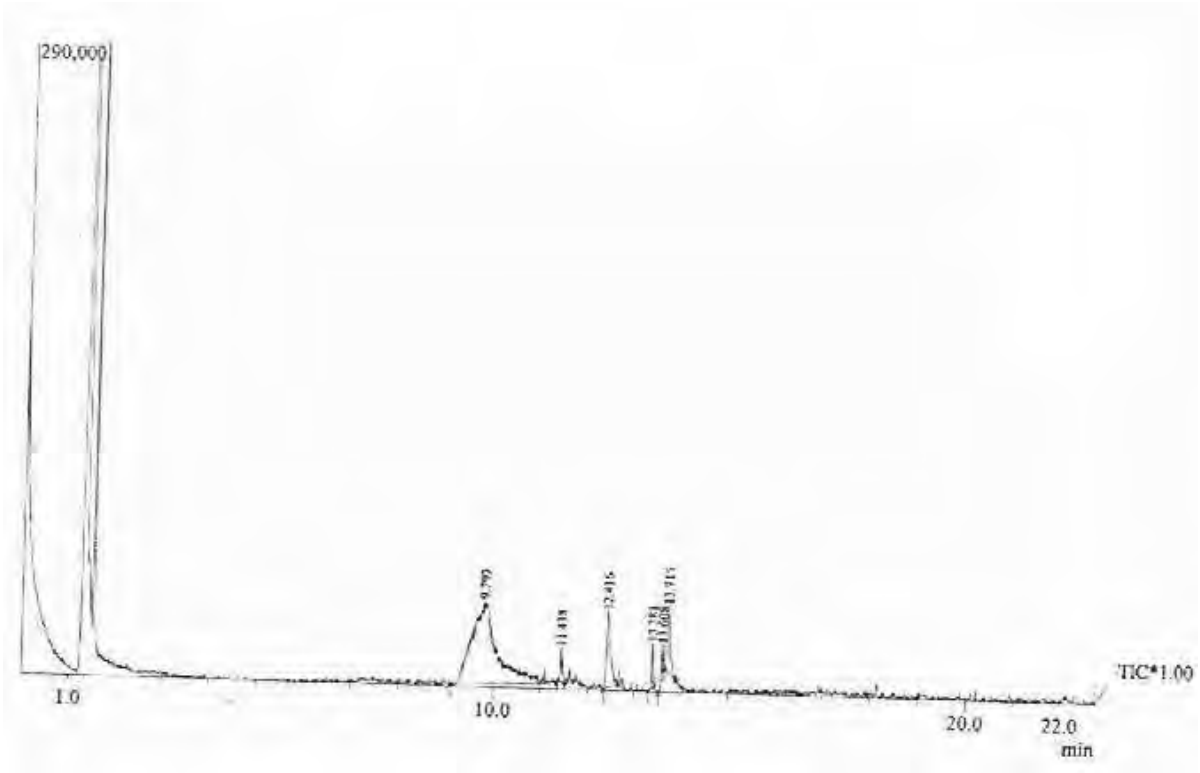

Fig. 3. Chromatogram of GC-MS analysis of biodegradation compounds

Table 2. The percentage of degradation of carbofuran to carbofuran phenyl in the mixed culture with the bacterium. Data represent average of 3 replicates ± SEM

Days after culture	Degradation (%)
1	02
3	23
5	51
7	64
9	64

followed by enhanced degradation up to 7 days of inoculation and then remained constant. However, it was observed, in total, the maximum degradation of cabofuran with the isolated bacterium was recorded as 64 per cent at *in vitro* cultures. Further, the degradation of the compound was confirmed by thin layer chromatography (TLC) and Gas chromatography and Mass Spectroscopy (GCMS) as shown in Figure. 3. Both the results indicated that carbofuran has been converted to crabofuran phenyl which is a less toxic substance.

CONCLUSION

The present work can be concluded that the isolated bacterial strain is a potential biodegrader of carbofuran compound. The further study of the bacterium will help us in establishing phylogenetic relationship among different strains of bacteria involved in the process of degradation and may further help us in development / transformation of other bacteria to acquire biodegradation process through recombinant DNA technology and to serves as new data base of microorganism with degradation capacity. Moreover, especially on the molecular characterization of the plasmid may help in developing certain strains of bacteria for bioremediation of carbamate group of pesticides *per se.*

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Mechanisation of cotton harvesting in India: Status, issues and challenges

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ABSTRACT : In India, efforts into mechanisation of cotton crops are being attempted for more than a decade. Numerous designs have been evaluated by Govt. and Private organisations. However, most of the designs failed to perform under field conditions. A commercial cotton picker prototype suitable for Indian cotton farms is being evaluated for harvesting of cotton crops sown on high density planting system for two cotton seasons. This prototype is developed by attaching the picker head of the worldwide used 6-rows cotton picker at the side of a 55 hp tractor with some specially designed attachments. The issues like cost of processing, yarn realisation percentage, fibre losses during cleaning operations, development of suitable genotypes, agronomic practices etc. that need to be addressed for successful mechanical harvesting of cotton crops in India are discussed in great length. The fibre parameters and trash analysis data pertaining to the field trials are also presented in this study. The field evaluation data and fibre quality indices reveal that the commercial picker to be launched for mechanical picking of cotton in India is promising.

Key words : Cotton harvesting mechanisation, cotton processing cost, defoliation, harvesting efficiency, losses, neps, Spindle picker, trash content

Cotton is a very important cash crop of India and it plays a leading role in the industrial and agricultural economy of the country. Cotton is main source of income in India for around 6 million farmers and about 40-50 million people are directly or indirectly engaged in cotton trade and processing [1]. India tops the world in cotton acreage and is the second only to China in cotton production with 35.1 million bales (*i.e.*170 kg each) production in 2012-13 [2]. Unlike the developed cotton growing countries (*i.e.* USA, Australia, Israel, etc.) where cotton is harvested using sophisticated machines called as cotton pickers/strippers, entire cotton in India is picked manually [3]. Moreover, there are around 28 cotton growing countries that harvest part of its cotton crop using cotton pickers/strippers. Cotton picking machines have spindles that pick (twist) the seed cotton from the opened cotton bolls (Fig. 1). The twisted seed cotton is doffed with the help of moist doffing pads wherefrom it is directed into a bucket attached at the top of the picker. Whereas cotton stripping machines use rollers equipped with alternating bats and brushes to knock the open bolls from plants to a conveyor (Fig. 1). The cotton picker plucks only the open bolls while the stripper strips both the open and the unopened bolls and some plant matters as well. Hence the stripped cotton contains more than two-three times trash content as compared to the machine picked cotton.

Though, the researches into cotton mechanisation in India started in around year 2000 under the NATP programme, the requirement of a suitable cotton mechanisation system has been felt very badly in last couple of years as the cost of cotton picking in India (i.e.

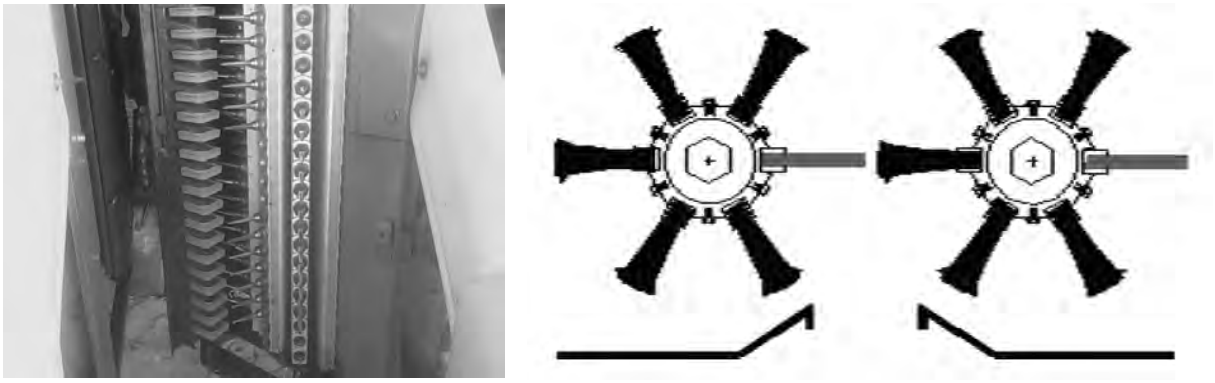

Fig. 1. Images of picker head (right) and stripper rolls with bats (left)

around Rs. 4-6/kg seed cotton) has doubled in recent past mainly due to high inflation rate, migration of landless farm labours to cities and implementation of the *National Rural Employment Guarantee Act* (*NREGA*). Moreover, the shortage of labours during peak season results in the delay in sowing of the next crop leading to low yield [4]. At present, the manual cotton picking cost in India is around 10-12 per cent of the total cotton selling price, which is much higher than the harvesting cost of any other crop grown in India. For instance, the rice harvesting cost is almost negligible compared to its selling price *i.e.* 0.01 per cent. In order to meet the scarcity of labour and to reduce the cotton picking cost, efforts need to be concentrated on mechanization of cotton harvesting [3].

In the last ten years, cotton acreage has been growing at an average annual rate of around 3%. However, the average cotton yield in India is steady at around 500 kg/ha compared to world average of 730 kg/ha (ICAC, 2010). The low yields of cotton in India are primarily due to rainfed cultivation, inadequate inputs usage, untimely field operations and inefficient crop production technologies. The lack of disease resistant and high yielding cotton varieties/ hybrids also contribute in low yield of cotton. Low cotton yield and increased cotton cultivation cost have reduced the farm income leading to a series of suicides committed by farmers in some cotton growing states of India. The mechanisation of cotton harvesting particularly in USA, Brazil, Turkey, etc. has led to significant increase in cotton productivity, decrease in cultivation cost and increase in farm income. Heinicke and Grove [5] have demonstrated positive effects on cotton yield by using machines for cotton harvesting in USA. Similarly, Isin*et al.,* [6] have also reported considerable increase in cotton yield in Turkey on adoption of mechanical picking. A recent study by Konduru *et al.,* [7] has estimated potential increase in cotton farm income in India *i.e.* around Rs. 10,000/ac, if cotton is harvested mechanically. The increased productivity in case of mechanical picking is mainly achieved by reducing row-row and plant-plant spacing which in turn increases the plant density by 4-5 times than the conventional method. The reduction in plant spacing is obtained by controlling the height and branches of the cotton plants.

Though, the farmers are in dire need for a suitable cotton harvester and it has potential for increasing productivity, farm income, availability of labours for other crops etc., there are many issues and challenges that need to be addressed for adoption of cotton harvesters in

India. Adoption of mechanical harvesting in India is not dependent upon just the availability of suitable harvesters. The successful adoption of cotton harvester in other part of world suggests the requirement of holistic change in the entire chain of cotton cultivation including breeding and agronomic practices, harvesting and processing operations for successful adoption of cotton pickers as all operations are interlinked. This paper explores the status, issues and challenges that require group efforts from scientists and technologists belonging to cotton breeding, agronomy, farm machinery, extension, cotton processing etc. for successful adoption of cotton harvesting and its economic feasibility.

Appropriate plant physiology : Manual crop harvesting, particularly manual cotton picking is mostly independent upon plant height, width, location of bolls on plants, etc. However, there are certain limitations in the functionality and capability of even highly sophisticated harvesting machines. It is required to suitably modify the plant physiology through genetic or breeding interventions to obtain a particular plant height, branch structure, locations of fruits on plants, etc. in order to successfully automate the harvesting operations. Mechanical cotton pickers require medium plant height *i.e.* around 1.0-1.2 m with minimum branches and bushes i.e. spreading into 0.5-0.6 m diameter for efficient and viable harvesting of cotton crops. Countries that employ mechanical cotton pickers have developed suitable plant genotypes having required plant physiology amenable for mechanical picking. Cotton varieties with the right plant architecture and height, amenable for mechanical harvesting need to be developed for mechanical pickers to work efficiently and effectively.

It is normal practice to sow about 25,000 to 40,000 plants/ac (two to three plants/foot of a row in conventional spaced rows: 38-40 inches) for picker type varieties [8] whereas 2 to 3 feet spacing/plant (*i.e.* 5000-6000 plants per acre) is the normal practice for sowing of *Bt* seeds in India. Though, the reduction of plant height and width results in less bolls/plant, the increased plant population per acre area results in more number of bolls than the convention method leading to increased productivity. Cotton breeders and scientists from government research organisations and private seed companies are working for past couple of years to develop cotton varieties, which are suitable for mechanical picking. M/S. Ankur Seeds Pvt. Ltd. and M/S. Nuzivedu Seeds Ltd. are working in tandem with M/S. John Deere India, a cotton picker manufacturer for development of suitable plant genotypes. In this cotton season, M/S. Ankur Seeds Pvt. Ltd. have evaluated its regular products, Ankur 8120 and Ankur 3028 hybrid *Bt* seeds for their suitability for mechanical pickers. It shows that instead of developing exclusive genotypes amenable for mechanical picking, most of the work is directed on identifying varieties from their regular products. The growth of the plants was regulated by using growth inhibitors or growth regulators developed by M/S. Bayer Crop Sciences, a German well known company for production of chemicals required for mechanical harvesting of cotton.

Synchronise boll opening : It is normal practice in India to harvest the cotton crops in 3-4 pickings because of occurrence of multiple flowering and fruiting of cotton that lead to development of 3-4 flushes of cotton bolls [9]. Though cotton pickers collect only fully open bolls and leave the unopened bolls on the plants unaffected, it's not economically viable to operate the mechanical picker more than once primarily

because of high diesel prices prevailing in the market. Delayed cotton pickings have also been attempted in earlier trials in order to allow unopened bolls to get matured. However, it did not work well as locules of bolls which were opened initially got unattached from its burrs and had fallen on the ground. Moreover, there is every chance for damage of opened bolls unpicked for a long time due to incessant rain and wind. There are some chemicals that are used to enhance the rate of boll openings. However, these chemicals affect the natural boll-opening process, but they do not cause bolls or fibre to mature faster. There is chance of affecting the maturity and micronaire values of fibres by improper timing and doses of chemicals. Hence, there are requirement for identification of proper chemicals, optimisation of its doses, timing etc.

Development of suitable cotton pickers : The average farm holding in India is less than 2 hectare and the size of Indian cotton fields is very small [10]. The machine pickers available in the world market are very large in size and capacity [11], hence they are unsuitable for cotton pickings in small Indian cotton farms. Dedicated work at different Indian Cotton Research Institutes and by Indian as well as foreign agricultural machinery manufacturers is going on for past one decade towards development of a suitable cotton picking machine for small sized Indian farms. Researchers have tried different picking methods (*i.e.* pneumatic suction, pneumatic suction cum picking brushes, sensor techniques, hand held picking machine, etc.) for harvesting of cotton [12]. However, most of these methods did not perform well under field conditions [13].

It has been observed by the numerous researchers that among the different methods tested for cotton picking, the conventional spindle type picker based mechanism appeared to be working satisfactorily for picking of cotton. This method was also evaluated in Indian cotton farms by cotton pickers imported from then USSR [14]. However, the further progress in this direction was constrained by the fall of former USSR.

Fig. 2. Single row cotton picker attempted in India

The potential for mechanical pickers in Indian market have attracted the global giants like John Deer and New Holland for development of mechanical pickers suitable for picking of cotton from small cotton fields. Efforts have been made by the researchers and agricultural machinery manufacturers for attaching the cotton picking heads in the side of existing tractors so as to avoid the high initial investment in purchasing a self-propelled spindle type picker. John Deer India has already come out with a single row cotton picker in which picker head is attached at the side of a 55 hp tractor (Fig. 2). This machine is being evaluated

for two cotton seasons at different part of the country along with several stakeholders including ICAR research institutes, state govt. officials, seed producing companies, chemical manufacturers, ginneries etc. [15]. New Holland is also carrying out the field testing of a cotton picker prototype specially designed for the Indian market expecting to be launched in 2-3 years' time. The proposed picker is tractor propelled and tailored specifically for the small farms suitable for Indian farmers.

Defoliation : Defoliation is the shedding of cotton leaves that naturally occurs when

Fig. 3. Cotton crop before defoliant application (left) and after defoliation (right)

leaves become physiologically mature (Fig.3). It is required to artificially shed the cotton leaves using certain chemicals called as defoliants or harvest aids in order to eliminate the main source of stain and trash to enter the cotton while harvesting. Defoliation also helps in improving lint grades, reduces moisture, improves storage of cotton and opens the green and unopened bolls [16]. There are a number of chemicals used for defoliation of cotton meant for mechanical harvesting [17]. The effectiveness of defoliation depends on several factors like temperature and rain fall at the time of treatment, periods of cloudy weather after treatment, soil moisture and nitrogen levels, calibration of application rates, etc. Weather conditions at the time of application and three to five days following application have a significant effect on cotton response to harvest aids. Harvest aids are most active when temperature, sunlight intensity and relative humidity are high. The yield and condition of the cotton crop are also deciding factor for the choice of defoliants. Optimisation and standardisation of defoliants still remain a challenge in India causing 4-5 per cent additional trash content in harvested cotton using mechanical picker in form of un-shedded leaves. It has also been observed in several cases where defoliation did not work properly due to certain unfavourable conditions resulting in 20-25 per cent trash content in harvested cotton against 10-12 per cent for properly defoliated cotton.

Field losses in terms of left over and fallen bolls : The cotton picker machine plucks the cotton from open bolls while unopened bolls are left over on the plants (Fig. 4). Moreover, some

part of the open bolls is also left un plucked on the plants and some part of harvested cotton falls on the field while harvesting. Field evaluation jointly conducted by CIRCOT, Nagpur, CICR, Nagpur and CIAE, Bhopal of a two row cotton picker (model no. 9935) imported from John Deere, USA during cotton seasons 2005-2006 revealed field losses to the tune of 10-15 per cent [18]. The issue of harvesting losses was also raised by group of farmers who witnessed the demonstration of cotton picker harvesting at Abohar, Punjab. The field loss due to mechanical harvesting of any crop is not a new phenomenon. Though combine harvesters also result in field losses, combine harvesting of wheat has become indispensible particularly in Northern part of India. The net margin of cotton farmers has bottomed out in recent past due to increased inputs and labour costs. However, benefits of cotton picker in terms of increased productivity by means of high plant density system and reduction of labour cost for picking have potential to offset the harvesting losses.

Fig. 4. Hand picking of cotton (left) and mechanical harvesting of cotton crop (right)

Requirement of additional pre-cleaning machinery : It is widely reported in literature [12, 14, 19-21] that the machine picked cotton contains around 10-15 per cent trash content, which includes burs, sticks, leaves, grasses, motes, etc. However, the imperfection of defoliants under Indian conditions has led to increase in trash content in mechanical picked cotton by 4-5 per cent. Moreover, trash content in range of 20-25 per cent was also observed in certain cases where defoliation did not work properly. The handpicked cotton particularly available in India hardly contains trash content in range of 2-5 per cent depending upon cotton varieties, skill of pickers, precautions taken during picking, number of flushes etc. There is requirement of a cylinder type pre cleaner developed by CIRCOT for pre cleaning of handpicked cotton prior to ginning. The cylinder type pre-cleaner removes around 25-30 per cent trash content from the cotton and the remaining trashes are removed in pneumatic conveying and lint cleaning operations. Finally, the bales processed from properly managed modern Indian ginneries contain around d"1 per cent trash content using a cylinder type pre cleaner and lint cleaner. On contrary, there is requirement of 3-4 number of additional special type pre cleaning machines based on combing and extracting principles for making the machine picked cotton ginnable. Cylinder cleaners use rotating spiked cylinders that open and clean the seed cotton by scrubbing it across a grid bars that allows the trash to sift through (Fig. 5). The

cylinder type pre-cleaners are meant for removal of kawadi/immature bolls, fine/pin trash, separation of metallic pieces and opening of the cotton. However, machine picked cotton contains large vegetative content like sticks and burrs and significant amount of green and dry leaves that require combing and extracting actions for dislodging of large and fine trashes. Large foreign matters are removed by combing action and centrifugal force in extractor type cleaners as seed cotton is pulled across a series of grid bars by a rotating saw/toothed cylinders. This cleaning mechanism is referred as the "sling-off" principle.

Fig. 5. Internal view of cylinder pre cleaner (left) and cylinder pre cleaner designed by CIRCOT and developed and marketed by BSI, Nagpur (right)

Cleaning machines based on sling off principles are not readily available in local market and import is unviable and very costly. Ginning Training Centre (GTC) of CIRCOT, Nagpur is working in tandem with the cotton picker research group towards development of a cleaning system suitable for pre-cleaning of the machine picked cotton. Research group at GTC of CIRCOT has reported bringing down trash content to around 4-6 per cent level from initial trash content of 12-14 per cent for machine picked cotton using set of cleaning machinery developed at GTC of CIRCOT under the NATP project. GTC of CIRCOT is also collaborating with M/s. Bajaj Steel Industries, Nagpur for optimisation and standardisation of cleaning machinery for processing of the machine picked cotton. However, formation of higher neps, reduction in staple length of fibres and spinning of fibres during pre cleaning etc. are some issues that need to be addressed before launching to the ginners.

Moreover, the machine picked cotton is likely to be wetter than handpicked cotton due to application of water for doffing of cotton in case of mechanical picking. The efficiency of pre-cleaning machines depends to considerable extent on moisture in cotton. There is requirement of optimum moisture content in cotton for pre cleaning machine to function effectively. The greater the moisture, lower the efficiency and *vice versa*. Hence, there is requirement of a tower drier system for bringing down moisture content in machine picked cotton to optimum level prior to pre cleaning.

Trash content and fibre parameters for the machine picked cotton : The assembly of machines displayed above have been evaluated jointly by CIRCOT, Nagpur and M/s. BSI, Nagpur

for its performance for cleaning of the machine picked cotton at Abohar, Punjab during ginning season 2013-2014 (Fig. 6). Table 1 and Table 2 depicts that the trash content in the machine picked cotton was 13.3 and 22.2 per cent for properly and improperly defoliated cottons, respectively. Hence it can be concluded that the defoliation plays a major role in deciding the trash content in the machine harvested cotton.

Fibre quality parameters analysed using HVI does not show any significant effect on fibre qualities for cotton variety Ankur 8120 after cleaning it in set of machines. However, significant differences in fibre parameters were observed for cotton variety Ankur 3028 after cleaning it in the set of machines. It is probably

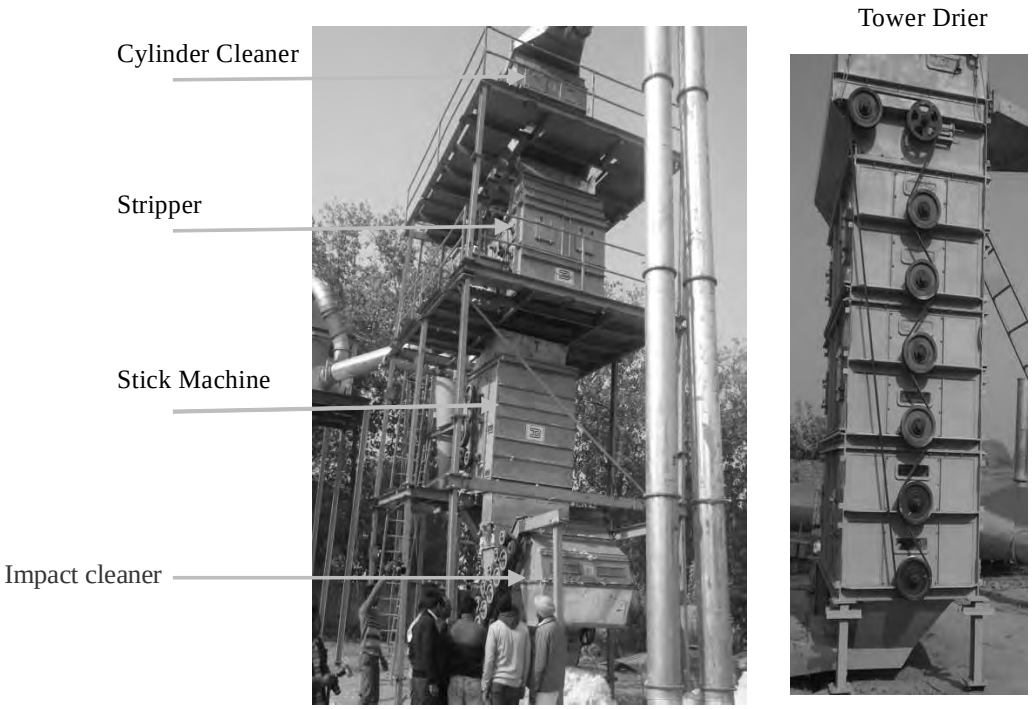

Fig. 6. Set of additional cleaning machines required for processing of the machine picked cotton (Courtesy: M/s. Bajaj Steel Industries, Nagpur)

Table 1. Trash content in properly defoliated cotton (Ankur 8120)

Seed cotton (g)	Lint (g)	Cotton seed (g)	Trash content	Bracts/ Burrs	Sticks	Green leaves	Immature seeds/ motes	Dry leaves	Total Trash content
300	104	196	(g)%	2.742.60	0.480.50	0.580.60	0.120.10	9.99.6	13.813.3

Table 2. Trash content in poorly defoliated cotton (Ankur 3028)

Seed cotton (g)	Lint (g)	Cotton seed (g)	Trash content	Bracts/ burrs	Sticks	Green leaves	Immature seeds/ motes	Dry leaves	Total trash content
300	104	196	(g)(%)	4.564.40	0.80.8	0.960.90	0.20.2	16.5815.90	23.122.2

Table 3. HVI results for machine picked cotton (MPC) and machine picked pre-cleaned cotton (MPPC)

Seed cotton	Average result upto 10 reading						
	UHML (mm)	ML (mm)	MIC (µg/inch)	Strength (g/tex)	UI (%)	SFI (%)	EL (%)
MPC A 8120	29.4	23.9	4.4	30.2	81.0	8.7	4.6
MPPC A 8120	29.4	23.6	4.3	29.6	80.1	8.7	4.7
MPC A 3028	28.1	22.8	4.9	29.1	81.2	8.9	4.5
MPPC A 3028	27.7	22.0	4.2	28.2	79.5	11.3	5.1

due to reason that the Ankur 3028 contained much higher amount of trash content than to Ankur 8120 that led to deterioration in the fibre qualities. Fibre parameters analysed using AFIS shows significant differences in the fibre parameters for both the cotton varieties tested in this study after pre cleaning (Table 4). Fibre neps that used to be around d"100 count/g for roller ginned Indian cotton have increased to around 250 count/g. Moreover, upper quartile length (UQL) measurements that corresponds to upper half mean length (UHML) of HVI values shows significant loss in fibre length (about 0.5 mm) for the pre-cleaned cottons. Higher neps count leads to reduction in yarn realisation and increase in fibre losses in spinning mills resulting in reduction in mill profits. The AFIS data clearly shows that the pre cleaning machines need to be optimised and fine-tuned in order to avoid damage in fibre qualities. Table 5 shows that the trash content in the machine picked cotton has been brought down to 4.2 per cent and 5.7 per cent, respectively for the properly defoliated and improperly defoliated cottons.

Increased investment for fixed and processing cost : As mentioned in the preceding section that the machine picked cotton requires 3-4 numbers of additional cleaners for its processing. The cost of additional pre cleaners including a tower drier and conveying system is around Rs. one crore at prevailing market rates for a ginning plant of around 10-15 bales/ha capacity. It is very difficult to convince the Indian ginners to invest additional one crore rupees in procurement of pre cleaning machinery meant for handling of the machine picked cotton.

Increased cost of processing : The additional cleaning machinery requires around 100 HP additional connected electrical load and

Table 4. AFIS results for machine picked cotton (MPC) and machine picked pre-cleaned cotton (MPPC)

Seed cotton	Length module test result					Nep module test result				
	L(w) mm	UQL (w) (mm)	SFC (w) (mm)	L (n) mm	SFC (n) % (mm)	5(%) L(n) (mm)	Fiber Neps Count /g	**Mean size (ìm)**	Seed coat Neps Count /g	**Mean size (ìm)**
MPC A# 8120	23.9	31.0	14.0	17.6	37.1	35.5	168	**661**	16	**1451**
MPPC A 8120	23.1	30.4	16.4	16.5	41.2	35.2	280	**666**	14	**1364**
MPC A 3028	20.9	27.1	18.6	15.5	42.1	31.6	206	**630**	10	**1214**
MPPC A 3028	19.7	25.7	21.4	14.5	45.5	29.6	242	**679**	15	**1172**

#Ankur

Table 5. Reduction of trash content in pre cleaning operations

Seed cotton	Initial trash content in the machine picked cotton (%)	Trash content in the pre cleaned cotton (%)	Reduction in the trash content (%)
Ankur 8120	13.3	4.2	68.4
Ankur 3028	22.2	5.7	74.3

a heating device for drying of the machine picked cotton in the tower drier. There is requirement of around 60 litre diesel and 60 unit electrical energy in an hour for running of the tower drier and the cleaning machinery resulting in expenditure of around Rs. 5000/h (including maintenance and operator cost) for pre cleaning and drying of the machine picked cotton. The increased cost on the processing comes to around Rs. 1/kg of seed cotton, which is around 67 per cent of the total cost of ginning of the handpicked cotton (around Rs. 1.5/kg seed cotton).

Increased losses of fibres in ginneries : It is well known fact that the ginners particularly in India are reluctant for employing even a light cylinder pre cleaner and post cleaner. It is mainly because of the reasons that the pre cleaners lead to losses of some fibres *i.e.* 1 per cent along with the removed trash content. However, most of the separated fibres are short fibres. The study by Arude *et al.,* [22] has showed processing losses to the tune of 1.8 and 3.3 per cent, respectively for handpicked cotton of the first and the second pickings. The application of 3-4 pre cleaners for the processing of the machine picked cotton shall lead to increased loss in fibres that may concern to the ginners.

CONCLUSIONS

This study presents an overview of status, issues and challenges for mechanisation of cotton harvesting in India. The following conclusions can be drawn from this work:

- Spindle type picker specially developed for harvesting of Indian cotton is found to be promising.
- It is required to develop suitable varieties/hybrids to obtain suitable plant types that are required for successful operation of the mechanical picker.
- There is requirement for adoption of certain agronomic practices like application of growth regulators, boll openers, defoliants etc. for successful operation of the mechanical picker.
- Trash content in the machine picked cotton was 13.3 per cent and 22.2 per cent for properly and improperly defoliated cottons, respectively.
- There is requirement of 3-4 additional cleaners based on sling off principles and a tower drier for processing of the machine picked cotton.
- Ginners shall have to make a substantial investment in machinery to get their gins properly equipped to process the machine picked cotton.
- The processing cost of the machine picked cotton is 67 per cent higher compared to the handpicked cotton.
- There is slight deterioration in the fibre parameters due to processing in additional cleaners.
- Spinners shall have to make some investment in machinery to process bales with higher neps content.

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Parameter estimation of pre harvest yield forecast models for cotton crop in Haryana

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Abstract : Zonal yield models incorporating a linear time-trend and weather variables each spanning successive fortnights within the growth period of cotton crop have been developed to predict the district level cotton yield(s) inHisar, Sirsa, Bhiwani, and fatehabad districts comprising the western zone of Haryana. Between year crop models have been developed within the framework of multiple linear regression analysis. Although the weather variables were statistically significant as predictors with reasonably high coefficients of determination (r^2), but the predictions had too high percent deviations to be acceptable and hence were deemed unsuitable for routine crop yield forecasting. To improve the predictive accuracy of the zonal-yield models, an independent variable in the form of Crop Condition Term (CCT) as categorical covariate was added with the weather variables. Inclusion of CCT as dummy regressors in the weather-yieldmodels significantly improved the accuracies of the district level yield predictions in the state. Alternatively, the crop yield models incorporating a number of biometrical characters spread over five-six successive stages within the growth period of cotton crop, have been developed to predict the yield(s) of American cotton hybrid/variety Pancham 541 and H 1236 and *desi* cotton variety HD-432 during the *kharif* 2013-2014 in Hisar district of the state. On the basis of developed models, the cotton yield of the selected hybrid/ varieties can be forecasted in the beginning of October by using biometrical characters total number of bolls and plant diameter.

Key words : DOA yield(s), dummy variable, linear time trend, opened bolls, per cent deviation, unopened bolls, weather variables, yield

The importance of agriculture for the Indian society can hardly be over emphasized, as its role in economy, employment, food security, self-reliance and general well-being does not need reiteration. India has a very well developed system for collection of crop statistics at village level and aggregating it at different administrative levels. However, the need for early and in-season crop production forecasting has been strongly felt. There has been substantial boost in food grain production over the years but it could not keep pace with rate of increase in the population of the country. Thus, increasing agricultural productivity has been main concern since the scope of increasing area under agriculture is rather limited. Fulfilling this requirement entails judicious planning based on information related to various aspects of agriculture. Thus, information on crop acreage, yield, production and conditions are important inputs for strategic planning.

Crop yield is affected by technological change and weather variability. It can be assumed that the technological factors will increase yield smoothly through time and therefore, year or some other parameter of time can be used to study the

overall effect of technology on yield. Weather variability both within and between seasons is the uncontrollable source of variability in yield. Weather variables affect the crop differently during differ-ent stages of development. Thus, the extent of weather influence on crop yield depends not only on the magnitude of weather variables but also on the distribution pattern of weather over the crop season which, as such, calls for the necessity of dividing the whole crop season into fine intervals. This increasesthe number of variables in the model and in turn, a large number of constants are to be evaluated from the long time-series data for precise estimation of the parameters.Thus, a technique based on relatively smaller number of manageable parameters and at the same time, taking care of entire weather distribution may solve the problem.

Various organizations in India and abroad are engaged in developing methodology for pre harvest forecast of crop yields using different approaches. Use of crop input and weathervariables forms one classof fore-casting crop yield. The other approach uses plant vigour meas-ured through plant characters. It can be assumed that plant characters are integrated effects of all the factors affecting crop yield. Yet another approach is measurement of crop vigour through remotely sensed data. The applications of Remote Sensing (RS) techniques for crop inventory have received attention right from the beginning in India. Among the various applications of remote sensing in natural resources management, its use for crop production forecasting is of great economic benefit. First attempt in the country towards the use of satellite digital data for crop acreage estimation was made in Karnal district of Haryana state using Landsat MSS data by Dadhwal and Parihar, 1985. Most commonly used models are based on regression approach. Broadly speaking, the two types of approaches are beingattempted *i.e.* Between year model and Within year model.

1.1 Between year models : These models are developed by taking previous years data. Objective yield forecasts are obtained by substi-tuting the current year data into a model developed on the basis of previous years. An assumption is made that the present year is a part of the composite population of the previous years.

1.2 Within year models : The 'between year models' while performing satisfactorily in typical years may fail in atypical years. A model which uses data from the current growing season only may be beneficial in improving forecasts during a year with atypical growing condi-tions. These models are developed to provide forecasts of pertinent components of crop yield relying entirely on growth data collected from plant observations during the current growing season. The model uses repeated observations from the current year to estimate the parameters needed to forecast the dependent variable at maturity.

Farmer is the best judge of the likely production in the fieldand thus the farmers' appraisal could serve as a good input for forecasting the crop yield. Expert opinion data may be collected in a number of rounds in a year by interviewing the selected farmers regarding their assessment about the likely crop production and chance of occurrences in various yield classes. A study has been carried out by Agrawal and Jain, 1996 to study the feasibility of using farmers' appraisal in the forecast model for sugarcane crop. The results revealed that a reliable forecast could be obtained using plant population and farmers' appraisal.

The Board of Agriculture in India recommended as early as 1918, an objective

method of conducting crop cutting experiments for estimation of crop yield through a random selection of villages, fields and plots. But the credit of carrying out the first yield estimation survey in 1923-1925 on the principles of random sampling goes to Hubback (1927). India has a well established system for collecting agricultural statistics. The primary responsibility for collection of data regarding the area and production under different crops is that of the State Governments. Yield estimates obtained through analysis of scientifically designed crop-cutting experiments (CCE) are routinely issued by the Directorate of Economics and Statistics. However, the final estimates by state Department of Agriculture (DOA) are given a few months after the actual harvest of the crop. Thus, one of the limitations of the conventional method used so far is timeliness and quality of the statistics. This makes these statistics unusable for planning and management purposes. Thus, developing an objective methodology for crop yield forecast models justify the need of such types of study.

India tops in terms of cultivated area (around one fourth of the world's acreage) and is the third largest producer of cotton in the world. Almost the entire cotton production is concentrated in nine major cotton growing states; Punjab, Haryana, Rajasthan, Gujarat, Maharashtra, Madhya Pradesh, Karnataka, Andhra Pradesh and Tamil Nadu. In Haryana State, Hisar and Sirsa are the two major cotton producing districts, accounting for 80 per cent of the acreage and 86 per cent of the cotton production in the state. Cotton is the dominant crop grown in these districts during *kharif* season and occupies almost 40 per cent of the geographical area. Cotton is mostly grown under irrigation due to the prevailing arid conditions. In Haryana state, the first attempt to estimate cotton acreage and condition assessment was made during *kharif,* 1990-1991 season in Hisar and Sirsa districts using IRS-1A LISS-1 digital data and the stratified random sampling approach (Sharma *et al.*, 1992). Subsequently, the efforts were made to improve the accuracy and timeliness of this process by modifying the stratification and sampling procedure by Yadav *et al.,* (1994). Larson *et al.*, (2002) have studied cotton defoliation and harvest timing effects on yield and quality. Viator *et al.*, (2005) have worked to observe the effect of climatic factors on cotton boll formulation. A study on the relationship between leaf area index and IRS LISS-III spectral vegetation indices of cotton in Hisar district was conducted by Kalubarme *et al.*, (2006).Verma *et al.,* (2012) have worked on pre harvest cotton yield forecasting based on plant biometrical characters in Hisar district.

2. Data description and modeling procedure adopted : Average yield statistics (1980-1981 to 2010-2011) of cotton for Hisar, Sirsa, Bhiwani and Fatehabad districts and the meteorological data on minimum temperature, maximum temperature, rainfall, sunshine hours and relative humidity for the same period werecollected for the purpose. Cotton crop is generally sown before the onset of monsoon (May-June) and is harvested during early winter (Nov.-Dec.). The daily weather data was converted into fortnightly data base over the cotton growth period *i.e.* 1stMay to 30th Novemberdivided into 14 fortnights. All the fourdistrictswere considered under the single zoneby considering weather parameters as regressors and state Department of Agriculture yield as regressand for the development of zonal yield models. The study has been categorized into two parts i.e. the development of Between-year and within year forecast models.

2.1 Between year cotton yield models using weather parameters and crop condition term as categorical covariate : Zonal yield models of cotton crop comprising ofHisar, Sirsa, Bhiwani and Fatehabad districts have been developed using the linear time-trend based yield and weather parameters computed over different fortnights of crop growth period. The models development/performance have been tried in alternative ways. First of all, the DOA yield data of cotton for Hisar, Sirsa, Bhiwani and Fatehabaddistricts were used by considering time (year) as an independent variable and had been regressed against yield to get the trend equation of the form $T_r = a+bt$; where T_r = trend yield (kg/ha), a = intercept, b = slope and t = year.

Model 1 :HisarBhiwaniSirsaFatehabad (district-level) Y= 288.29+ 4.20*t Y= 254.34+ 3.14*t Y= 287.32+ 7.26*t Y= 180.45+53.04*t (R^2= 0. 27, n=44) (R^2= 0.19, n=37) (R^2= 0.33, n=35) (R^2= 0.74, n=10) (Insufficient data)

Further, the linear trend based yield(s) along with time-series weather data on minimum temperature, maximum temperature and rainfallwere used for the development of regression models at zonal level. The general linear regression model used is defined as follows:

$$Y = a_0 + \sum_{i=1}^{14} b_i TMX_i + \sum_{j=1}^{14} bjTMNj + \sum_{k=1}^{14} b_k ARF_k + cT_r + e$$

where, Y = Cotton yield (kg/ha)

a_0 = Intercept

b_i, b_j, b_k = Regression coefficients of weather parameters (i,j,k - meteorological fortnights)

c =Regression coefficient of trend yield

TMX=Average maximum temperature

TMN= Average minimum temperature

ARF= Accumulated rainfall

T_r= Trend yield (kg/ha)

e = the error term with assumption NID $(0, s^2)$

The multiple linear regression analysis for various combinations of trend-agromet variables was carried out. Crop yield modelswere selected using the stepwise regression programme (Draper and Smith, 2003) in which the variables were included or excluded one at a time with decisions at any particular step conditioned by the previous step. The weather-yield relationships selected on the basis of highest adjusted R^2 and lowest standard error of estimate (SE) are described below:

Model 2

$Yield_{est}$ =699.20+.184 Tr+5.20 TMX_{sep2}-20.41TMN_{july2}-1.83ARF_{may2}-0.89ARF_{jun1}

R^2 = 0.45, adj. R^2 = 0.37 & SE = 64.8

The weather parameters were turned out to be the significant predictor variables but couldn't solve the purpose of crop yield prediction and the relative deviations from DOA yields were beyond tolerable limits for the sample period itself and thus leaving the above fitted models unsuitable for crop yield forecasting purpose.Seeing the performance of the developed models, the need was felt to insert more weather parameters. So, the stepwise regression analysis was again performed for deciding the zonal model by including relative humidity and sunshine hours. Once again, the fitted models couldn't provide the satisfactory response for pre-harvest crop yield estimation purpose. Alternatively, one more regressor *i.e.* crop condition term (CCT) was generated as categorical covariate on the basis ofDOA crop yield series. Proceeding further, the above mentioned weather parameters spread over 12 fortnights along with CCT as dummy

variable were used for obtaining the zonal yield models. The results obtained in view of the above possibilities are expressed below:

2.11 Use of categorical variable along with weather parameters

Model 3

$Yield_{est} = 315.39 + 13.81SSH_{oct2} - 7.39TMN_{aug1} - 0.18 ARF_{jul2} + 169.24D_1 + 348.60D_2$

$R^2 = 0.80$, adj. $R^2 = 0.76$& SE= 53.58

Model 4

$Yield_{est} = 242.30 + 0.11\ Tr + 15.34\ SSH_{oct2} - 6.82\ TMN_{aug1} - 0.18\ ARF_{jul2} + 169.68D_1 + 343.79D_2$

$R^2 = 0.80$, adj. $R^2 = 0.73$& SE= 54.69

Table 1. District-specific estimated cotton yield(s) (Est. yield) based on zonal model and their associated percentage deviations (RD (%)= 100 ´ (Est. yield-observed yield)/ observed yield)

District	Year	DOA yield (kg/ha)	RD(%)
Bhiwani	2009-2010	455	-8.54
	2010-2011	433	9.26
Hisar	2009-2010	610	-3.58
	2010-2011	497	4.34
Fatehabad	2009-2010	745	-6.18
	2010-2011	508	5.87
Sirsa	2009-2010	706	-3.88
	2010-2011	558	12.07

Here, may_1, may_2, jun_1, jun_2,.....nov_1, nov_2 refer to different fortnights and D_1, D_2 are dummy variables i.e. one less than the number of three categories of CCT prepared on the basis of trend based yield .

2.2 Within year models based on plant biometrical characters

Study area, sampling plan and recording of the observations : The study was undertaken in the cotton section area of Genetics and Plant Breeding, CCS HAU, Hisar for collecting primary data on biometrical characters.The Hisar district, a part of the Indo-Gangetic alluvial plain is situated between $28^0$53'45" to $29^0$49'15"N latitudes and $75^0$13'15" to $76^0$18'15"E longitudes. It occupies an area of 3788 sq km. Most of the total irrigated area in the district is under canal irrigation because of brackish underground water. The Hisar district experiences a sub tropical climate. The climate is influenced by westerly winds in summer months raising temperature as high as 48^0C, whereas in winter north westerly cold winds provide low temperature touching even 0^0C. The average rainfall in the district is 334.4 mm. About 85 per cent of annual rainfall is received during the south western monsoon period.

The primary data on biometrical characters *i.e.* plant height, plant diameter, number of opened and unopened bolls, boll size, flower and boll shedding,fruiting points available and yields at various pickings were collected during 2013-2014 for developing pre/harvest crop yieldforecast models. Thirty plantseach of American hybrid/varieties Pancham 541, H 1236 and *desi* cotton variety HD 432 were selected for recording of observations on biometrical characters.These plants were tagged and the recordings were made at regular interval of a fortnight. The row to row spacing was 1m for all the three hybrid/varieties and plant to plant spacing was 45 cm for Pancham 541 and 30 cm for H 1236 and HD 432. The data were recorded from the first week of September to the second week of November for Pancham 541 and H 1236. For HD 432, the data were taken from the last week of August to second week of November. The observations on biometrical characters for each selected plant during different stages of growthfor all the threehybrid/varieties are

shown below:

Regression models via step-wise regression have been fitted, considering plant biometrical characters as regressors and total

Pancham 541 and H 1236

Stage I (1st week of September)

X_1 Height(cm)
X_2 Diameter (cm)
X_3 Unopened bolls

Stage II (3rd week of September)

X_4Height(cm)
X_5Diameter (cm)
X_6Unopened bolls
X_7Opened bolls
X_8Total bolls

Stage III (1st week of October)

X_9Height
X_{10}Diameter (cm)
X_{11} Unopened bolls
X_{12}Opened bolls
X_{13}Total bolls
X_{14} Boll size
X_{15}Flower and boll shedding
X_{16}Fruiting points available
X_{17} Yield of 1st pick (g)

Stage IV (3rd week of October)

X_{18}Unopened bolls
X_{19}Opened bolls
X_{20}Total bolls
X_{21}Yield of 2ndpick (g)
X_{22}Yield of (1st+ 2nd)picks (g)

Stage V (1st week of November)

X_{23} Unopened bolls
X_{24} Opened bolls
X_{25}Total bolls
X_{26}Yield of 3rdpick (g)
X_{27} Yield of (1st+2nd+3rd)picks (g)

Stage VI (2nd week of November)

X_{28}Unopened bolls
X_{29}Opened bolls
X_{30}Total bolls
X_{31}Yield of 4thpick (g)
Y Total yield (g)

HD 432

Stage I (Last week of August)

X_1 Height(cm)
X_2 Diameter (cm)
X_3 Unopened bolls

Stage II(2nd week of September)

X_4 Height(cm)
X_5Diameter (cm)
X_6 Unopened bolls

Stage III (Last week of September)

X_7Height(cm)X_8Diameter (cm)
X_9Unopened bolls
X_{10} Opened bolls X_{11}Total bolls
X_{12} Boll sizeX_{13}Flower and boll shedding
X_{14}Fruiting points available
X_{15} Yield of 1st pick (g)

Stage IV (2nd week of October)

X_{16}Unopened bolls
X_{17} Opened bolls
X_{18}Total bolls
X_{19}Yield of 2nd pick (g)
X_{20}Yield of (1st+ 2nd)picks (g)

Stage V (Last week of October)

X_{21}Unopened bollsX_{22}Opened bolls
X_{23} Total bolls
X_{24}Yield of 3rd pick (g)
X_{25}Yield of (1st+2nd+ 3rd)picks (g)

Stage VI (2nd week of November)

X_{26} Unopened bolls
X_{27} Opened bolls
X_{28} Total bolls
X_{29}Yield of 4thpick (g)
Y Total yield (g)

yield (Y) as dependent variable. The analysis was performed to develop suitable crop yield models in respect of cotton hybrid/varieties.

The selected yield forecast models based on plant biometrical charactersfor Pancham 541, H-1236 and HD-432 are:

Pancham : Yield = -47.46 + 3.02 total bolls + 34.89dia

R^2 = 0.81, adj. R^2 = 0.80 and SE = 18.13

H 1236 : Yield = -57.15 + 1.52 total bolls + 46.31dia

R^2 = 0.71, adj. R^2 = 0.69 and SE = 16.15

HD 432 : Yield = 8.85 + 2.31total bolls

R^2 = 0.73, adj. R^2 = 0.72 and SE =16.34

The yield of Americancotton hybrid/ varietyPancham 541 and H 1236 can be forecasted in the first week of October using the biometrical characters total bolls and plant diameter. The estimated yield based on the selected models were 18.33q/ha and 18.03q/ha respectively. Total number of bollswas the main contributing variable towards yield for *desi* cotton varietyHD-432 and the yield can be forecasted in the last week of September. The estimated yieldon the basis of fitted model was 19.43 q/ha.

Models based on plant biometrical characters at different stages of the cotton hybrid/ varieties

Hybrid/variety	Stage	Fitted Equation	R^2	Adj R^2	SE
Pancham 541	I	Y = -84.83 + 2.22 ht	0.53	0.52	28.10
	II	Y= -113.10 + 64.63dia +0.88ht	0.61	0.59	25.99
	III	Y = -47.46 + 3.02 total bolls + 34.89 dia	0.81	0.80	18.13
H 1236	I	Y = -96.06+88.03dia	0.55	0.53	19.87
	II	Y= -81.92+66.96dia+1.58totalbolls	0.63	0.60	18.39
	III	Y = -57.15 + 1.52 total bolls + 46.31dia	0.71	0.69	16.15
HD 432	I	Y = -32.38 + 55.08dia	0.32	0.29	26.04
	II	Y = -41.08 +5.27 unopened bolls +35.39 dia	0.67	0.65	18.42
	III	Y = 8.85 + 2.31 total bolls	0.73	0.72	16.34

DISCUSSION

Multiple linear regression models incorporating linear time-trend and weather variables representing successive fortnights within the cotton crop growth period have been developed to predict district-level cotton yield(s) in western zoneof Haryana. Initially, the weather models were developed using three weather parameters *viz.*, maximum temperature, minimum temperature and rainfall keeping in mind to have the crop yield estimates in the beginning of October. However, the zonal weather model obtained to provide the crop yield estimates in the beginning of October was not capable of capturing sufficient variability (adj. R^2 = 0.37). Further, more weather variables were exhausted to assess the maximum contribution of the climatic regressorsspread up to 2nd fortnight of October. Several weather variables were identified as significant predictors of cotton yield. Adding linear time-trend to the model with the selected weather variables did not significantly improve the accuracy of the yield predictions. The predicted yields based on the models incorporating the weather variables selectedby stepwise regression analysis had rather wide per cent

deviations from the DOA yield estimates, sometimes wider than acceptable limits.Hence, the weather –yield models were again found unusable for yield forecasting purpose.

Seeing the performance of weather models, we attempted to improve the predictive accuracy of the models by identifying and adding additional covariate to the model with the selected weather variables. In particular, adding a crop condition term (categorical variable) to the models with the selected weather variables substantially improved the predictive accuracy of the models and produced what we consider to be quite satisfactory district level yield predictions using model 3.It is worth emphasizing that the weather variables and linear time-trend we considered alone may not suffice as a basis for accurate forecasting of cotton yield. It is therefore useful to explore and identify additional suitable agronomic/ biometrical variables that may further enhance the predictive accuracies of the models as we did with CCT for the district level yield predictions.

Secondly, within year cotton yield models based on plant biometrical characters were fitted. On the basis of developed models, the cotton yield of the selected hybrid/varieties can be forecasted in the beginning of October by using biometrical characterstotal number of bolls and plant diameter.

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Positive discrimination in *Bt* cotton and non *Bt* cotton cultivation in Karnataka: An economic analysis

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Cotton (*Gossypium* sps) the "White Gold" and "King of Fibers" is cultivated in 70 countries of the world and enjoys a predominant position amongst all cash crops in the world. The major producers of cotton are China, India, USA, Pakistan, Uzbekistan, Argentina, Australia, Burkina, Turkmenistan, Mali, Brazil, Mexico, Turkey and Egypt. These countries production is estimated around 119.243 million bales of 480 lbs in 2014-2015.India has become the leading producer of cotton and also maintaining the largest area under cotton and second largest exporter of cotton next to United States. India also sustained the position of being the second largest consumer of cotton and is expected to consume 24 million bales in 2014-2015 (Anonymous, 2014).

Bt (*Bacillus thuringiensis*) cotton was first genetically modified (GM) crop approved in India during 2002 for commercial cultivation. The farming community has widely adopted this technology at un precedented rate in agriculture. Hence, this is the fastest diffusion of any new crop technology in the history of mankind. This is mainly due to reduction of production cost, insecticide application (40-60%) and lessened insecticides spraying from 24 to 2-3 sprays/ season. Further, the area under *Bt* cotton has increased from 50,000 ha in 2002 to 11.60 million ha in 2014, a 230 fold increase equivalent to 95 per cent of the total cotton crop coverage (12.25 million ha) in the country. *Bt* cotton has contributed to increase the production of cotton by three times from 13 million bales to 40 billion bales during 2002 to 2014 and a target of 100 billion bales by 2030. This is mainly due to the introduction of *Bt* technology which spurred hybridization of cotton from three hybrids in 2002 to 1167 in 2014. *Bt* cotton transformed India from a net importer to a net exporter of cotton. India's cotton export registered to sharp increase from 0.5 million bales in 2001-2002 to 11.4 million bales in 2013-2014. (www.isaa.org).

In India, the major cotton growing states are Maharashtra, Gujarat, Andhra Pradesh, Madya Pradesh, Rajasthan, Haryana, Punjab, Karnataka and Tamil Nadu. In Karnataka, *Bt* cotton occupies an area of 7.60 lakh ha with a production of 26.90 lakh bales with a productivity of 626 kg/ha. The increase in productivity from 247 kg/ha in 2005-2006 to 626 kg/ha in 2014-2015 was mainly due to cultivation of *Bt* cotton (Anonymous, 2014). The predominant *Bt* cotton growing districts are: Dharwad, Gadag, Kalaburagi, Haveri, Raichur, Belagavi, Davangere, Bellary, Mysuru and Chamarajanagara.

The commercial value and importance of *Bt* cotton with respect to its area and production is gaining importance in the country at large and more so in Karnataka in recent years. Therefore, an attempt has been made in the present paper to review the empirical evidences on knowledge, adoption, economic performance, perception about benefits, risks

and superiority, cost returns besides suggestions of *Bt* cotton growers compared to non *Bt* cotton growers.

Empirical evidences : Studies conducted at the University of Agricultural Sciences, Bengaluru. (UASB) relevant to economic aspects of Bt cotton and non- Bt cotton growers have been reviewed. The important findings of studies were as follows:

A) Knowledge, adoption and economic performance of *Bt* cotton growers : Pruthvi (2011) conducted a study on farmers' knowledge and perception about *Bt* cotton with a sample of 60 farmers selected from six villages in Davanagere district. Laxmi (2012) conducted a study with the main objective of analyzing adoption level and economic performance of 120 *Bt* cotton growers in eight villages selected from two taluks in Gadag district. Similarly, Rahul Pawar (2015) studied the economic performance of Bt cotton growers in two taluks of Belagavi district.

The findings of three studies indicates that majority of the farmers had high knowledge (35.00%) and low (45.00) to medium (40.00%) adoption respectively. With respect to economic performance, majority of them had medium level in two studies (35.83% and 63.33%).This implies that farmers are interested in cultivation of Bt cotton in Karnataka as knowledge is a decisive factor of adoption and determinant of production.

(B) Farmers' perception about benefits of cultivating *Bt* cotton compared to non *Bt* cotton : Two studies conducted to relevant to farmers' perception about benefits of cultivating *Bt* cotton compared to non *Bt* cotton growers. The important findings of studies were as follows:

Majority of farmers perceived that the *Bt* cotton requires less number of sprays than non *Bt* cotton (2.97 score) followed by two statements "*Bt* cotton is compatible with existing farming practices of the locality" (2.88 score), "*Bt* cotton fetches higher price in market than non *Bt* cotton" (2.88 score) and also the "*Bt* cotton adoption will increase the income" (2.84 scores). Similarly, "*Bt* cotton fetches higher price in market than non *Bt* cotton" and "*Bt* cotton cultivation is easier than conventional cotton" statements shared the equal scores (2.78).

Majority of respondents perceived that "*Bt* cotton seed cost was very high compared to non *Bt* cotton" (2.97 and 2.92 scores) followed by "More premature drying of bolls" (2.85 and 2.53scores), "Difficult to get *Bt* cotton seeds in time" (2.54 score) and "*Bt* cotton was susceptible to moisture stress and drought" (2.50 score) as major risks in *Bt* cotton cultivation.

Cent per cent of the respondents have perceived that there was a reduction in the boll worm incidence and number of chemical sprayings by adopting *Bt* cotton (in both the studies) Whereas, reduction in sucking pests incidence was observed in one study. Further, majority of them perceived that there was a

Table 1. Knowledge, adoption and economic performance of *Bt* cotton growers

Categories	Knowledge	Adoption		Economic performance	
	Pruthvi(2011) (n=60)(%)	Pruthvi(2011) (n=60)(%)	Laxmi (2012) (n=120)(%)	Laxmi (2012) (n=120)(%)	Rahul Pawar (2015) (n=120)(%)
Low	33.33	21.67	45.00	33.33	22.50
Medium	31.67	40.00	25.83	35.83	63.33
High	35.00	38.33	29.17	30.84	14.12

Table 2. Farmers' perception about benefits of cultivating *Bt* cotton compared to non *Bt* cotton

Sl. No.	Statements	Pruthvi (2011) (n=60) Meanscore	Laxmi (2012) (n=120) Meanscore
1	*Bt* cotton cultivation is easier than conventional cotton	2.55	2.78
2	*Bt* cotton is compatible with existing farming practices of the locality	2.88	2.40
3	*Bt* cotton yields more than the conventional cotton	1.58	2.67
4	*Bt* cotton reduces costs on insecticides	2.50	2.67
5	*Bt* cotton quality is good compared to non *Bt* cotton	2.07	2.46
6	Saves labour costs than non *Bt* cotton	2.03	1.75
7	Requires less number of sprays than non *Bt* cotton	2.97	2.16
8	*Bt* cotton is more profitable than non *Bt* cotton	1.87	1.83
9	*Bt* cotton reduces reliance on credit	1.22	2.17
10	*Bt* cotton fetches higher price in market than non *Bt* cotton	2.78	2.88
11	Pest incidence is less than non *Bt* cotton	2.67	2.39
12	Environmental safety is possible through cultivation of *Bt* cotton	2.67	2.24
13	*Bt* cotton adoption will increase the income	-	2.84

reduction in the cost incurred towards pesticide application (94.62% and 91.67%), cost of production (90.04% and 81.67%), labour cost (92.17% and 80.00%) and disease incidence (87.98 %). Similarly, majority of respondents perceived increased in Bt cotton cultivation yield (91.83%) and increase in market price (88.81% and 65.00%) as superiority parameters

Table 3. Farmers' perception about risks of cultivating *Bt* cotton compared to non *Bt* cotton

Sl. No.	Risk statements	Pruthvi (2011) (n=60) MeanScore	Laxmi (2012) (n=120) MeanScore
1	Premature drying of bolls is more	2.53	2.58
2	*Bt* cotton is susceptible to moisture stress and drought	1.85	2.50
3	*Bt* cotton plants are brittle and the branches gets broken easily	1.03	1.88
4	*Bt* cotton is more susceptible to sucking pests	1.55	1.73
5	*Bt* cotton staple length is less than conventional cotton	1.18	1.57
6	*Kapas picking* in *Bt* cotton is more labour consuming	1.50	1.93
7	*Bt* cotton price is less than non *Bt* cotton	1.08	1.26
8	*Bt* cotton average weight of bolls is less than non *Bt* cotton	2.03	1.62
9	*Bt* cotton seed cost is very high compared to non *Bt* cotton seeds	2.97	2.92
10	*Bt* cotton is not suitable for rain fed condition	-	2.77
11	It is difficult to get *Bt* cotton seeds in time	1.82	2.54
12	*Out crossing* of *Bt* cotton pollen may lead to development of gigantic plants and weeds	1.15	2.17
13	*Bt* cotton cultivars requires more irrigation and fertilizers application	1.03	1.93
14	*Bt* cotton is not suitable for rainfed condition	1.07	-

Table 4. Farmers' perception about superiority of *Bt* cotton compared to non *Bt* cotton

Sl. No.	Parameters	Pruthvi (2011) (n=60)		Laxmi (2012) (n=120)		
		Increased (%)	No difference (%)	Decreased (%)	Increased (%)	Decreased (%)
1	Boll worm incidence	0.00	0.00	100.00	0	100
2	Sucking pests incidence	16.67	60.00	23.33	0	100
3	Disease incidence	3.33	73.33	23.33	11.62	87.98
4	Number of plant protection chemical spraying	0.00	0.00	100.00	0	100
5	Pesticide cost	0.00	8.33	91.67	4.98	94.62
6	Labour cost	0.00	20.00	80.00	92.17	7.42
7	*Bt* cotton yield over conventional cotton	33.33	13.33	53.33	91.83	8.3
8	Cost of production	8.33	10.00	81.67	5.81	90.4
9	Market price	65.00	26.67	8.33	88.81	10.79

Bt cotton growers were of the suggestion that providing good quality seeds (73.61% and 71.67%) followed by timely information about production practices (68.33 % and 43.61%), fixing minimum support price (67.67% and 60.00 %) and reduction of seed price (60.00 % and 56.94%) as suggestions to promote *Bt* cotton cultivation in the region. This calls for strengthening of extension services in the region to promote Bt cotton cultivation. As Mosher (1966) cites " the accelerator of agriculture development include providing quality seed (input), timely information (input) and support price besides, marketing niche"

(C) Costs and returns in *Bt* cotton and non *Bt* cotton : Manjunath Kerur (2012) has investigated the economics of *Bt* and non *Bt* cotton cultivation in five villages of Hirekerur taluk in Haveri district. The major findings of the study were as follows:

The total cost of cultivation of *Bt* cotton and non *Bt* cotton was in the order of Rs. 16.642 and Rs 17, 549 respectively. The cost of cultivation of non *Bt* cotton was found to be higher by Rs. 907 as compared to with that of *Bt* cotton. Human labour was the major component accounting for 24.26 per cent and 24.04 per cent in *Bt* cotton and non *Bt* cotton, respectively. The cost of bullock / machine labour, fertilizers and plant protection chemicals was higher in case of non *Bt* cotton cultivation at Rs. 2,024, Rs. 1,780, and Rs 2788/ac, respectively as compared to *Bt* cotton cultivation Rs. 1,516, Rs. 1,624, and

Table 5. Suggestions offered by the *Bt* cotton growers

Suggestions	Pruthvi (2011) (n=60)(%)	Laxmi(2012) (n=120)(%)
Provide good quality seeds	71.67	73.61
Reduce seed price	60.00	56.94
Fixing minimum support price	60.00	67.67
Provide timely information about production practices	68.33	43.61

Table 6. Comparison of cost-benefit ratio of *Bt* cotton and non *Bt* cotton (Rs/ac)

Sl. No.	Items	*Bt* cotton	Non *Bt* cotton
1	Seeds	1410	482
2	Human labour	3280	3233
3	Machine labour	1175	1175
4	Bullock labour	1457	1309
5	Farm yard manuare	1607	1359
6	Chemical fertilizers	3335	3153
7	Plant protection Chemicals	2301	3934
8	Irrigation	1173	1131
9	Picking	5523	4459
10	Marketing cost	541	404
11	Total cost	21075	19987
12	Yield (q/ac)	10.83	8.09
13	Gross return	41993	30516
14	Net return (C-A)	20914	10579
15	Cost Benefit Ratio	1:1.98	1:1.52

Source: Manjunath Kerur (2012)

Rs. 1,580/ac, respectively in that order.

The cost of seeds/ac was higher in case of *Bt* cotton (Rs.1897) compared to non *Bt* cotton cultivation (Rs. 1048). Similarly, the marketing cost was higher in case of *Bt* cotton (Rs.845) compared to non *Bt* cotton (Rs. 713). The proportion of fixed cost was found to be higher in case of *Bt* cotton producing farms (Rs 4049) compared to non *Bt* cotton producing farms (Rs.3634) it is imperative to note that input costs of (seeds) marketing cost were found to be more in *Bt* cotton compare to non *Bt* cotton which means production cost and marketing cost were found to be higher in *Bt* cotton cultivation over the non *Bt* cotton.

The /ac average yield of *Bt* cotton (9.15 q) was higher than that of non *Bt* cotton (6.84 q). The returns structure of *Bt* cotton revealed that gross returns was higher (Rs. 32, 025) in Bt cotton as compared to that of non *Bt* cotton (Rs. 23,940).

Table 7. Economics of *Bt* and non *Bt* cotton cultivation (Rs/ac)

Sl. No.	Particulars	*Bt* cotton		Non *Bt* cotton	
		Value	Per cent value	Value	Per cent value
A	Variable cost				
1	Seeds	1897	11.39	1408	8.02
2	FYM	696	4.18	536	3.05
3	Human labour	4038	24.26	4220	24.04
4	Bullock/machine labour	1516	9.1	2024	11.53
5	Chemical fertilizers	1624	9.75	1780	10.19
6	Plant protection chemicals	1580	9.53	2788	15.88
7	Interest on working capital @ 7 per cent/annum	397	2.38	446	2.54
	Total variable cost (TVC)	**11748**	**70.59**	**13202**	**75.25**
B	**Fixed cost**				
1	Depreciation	715	4.18	685	3.9
2	Land revenue	50	0.3	50	0.28
3	Rental value of land	1870	11.37	1870	10.65
4	Interest on fixed assets @11 per cent/annum	1414	8.49	1029	5.86
	Total fixed cost (TFC)	**4049**	**24.34**	**3634**	**20.69**
C	**Marketing cost**	**845**	**5.07**	**713**	**4.06**
	Total cost of cultivation	**16642**	**100**	**17549**	**100**

Source: Manjunath Kerur (2012)

Table 8. Yield and returns in *Bt* and non *Bt* cotton cultivation (Rs/ac)

Sl. No	Particulars	*Bt* cotton	Non *Bt* cotton
1	Yield (q)	9.15	6.84
2	Return		
	Main product	32025	23940
	Byproduct	170	212
	Gross returns	32195	24152
3	Total cost	16642	17549
4	Net returns	15553	6603
5	Total variable cost	11748	13202
6	Returns over variable cost	20447	10950
7	Returns/rupee of expenditure	1.93	1.37

The net returns of Bt cotton (Rs. 15,553) was higher than non *Bt* cotton (Rs. 6,603). The returns over variable costs was more in case of *Bt* cotton (Rs. 20,447) as compared to non *Bt* cotton (Rs. 10,950). Returns/rupee of expenditure was also higher in *Bt* cotton (Rs. 1.93) when compared to non *Bt* cotton (Rs.1.37) Table 8 gives more details of this fact.

CONCLUSION

From the fore mentioned studies it is observed that the Bt cotton growers had high knowledge, medium to low adoption and medium economic performance. Majority of respondents perceived that less number of sprays, less pest incidence, environmentally safe, compatible with existing farming practices and more income as important benefits of *Bt* cotton. With regard to the risks, more seeds cost, difficult to get seeds in time, more premature dropping of bolls and susceptibility to moisture stress and drought were perceived by majority of the respondents. Further, higher returns and increased income in Bt cotton demonstrated the profitability of the crop. Hence, it is inferred that organizing extension educational activities such as trainings, demonstrations, seminars, exhibitions, field days, field visits etc., is the need of the hour to enhance the knowledge and adoption of *Bt* cotton growers. The scientists of public and private sectors should try to develop new *Bt* cotton hybrids to overcome the risks. Further, quality seeds be made available in time in sufficient quantity by the concerned organizations to achieve sustainability in Bt cotton production in the ensuing years.

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Role of extension agencies in increasing the cotton production in India

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Abstract : India is one of the important contributors of world cotton production and cotton plays a significant role in Indian economy. Over the years the area, production and productivity of the crop has undergone significant changes. Many factors were responsible for the desirable changes occurred in the production level of cotton in the country. Among them, the efforts taken by the various cotton extension agencies and the effectiveness of the cotton extension programs executed are very vital to document. The establishment of various institutions in India in 20th and 21st centuries accelerated the outreach programs of cotton. The first line extension programs of Indian Council of Agricultural Research facilitated the effective transfer of latest cotton technologies. Among the various first line extension programs of ICAR, the FLDs a typical kind of national demonstrations played a major role in disseminating novel cotton production and protection technologies to cotton growers. The Technology Mission on Cotton with the objective of improving production, productivity and quality of cotton by the GOI enabled Indian cotton to compete globally. The recent approach "e-*Kapas* network", an ICT initiative and mobile phone based extension model facilitated the cotton researchers to reach the unreached in a swift manner. Analysis on all these efforts revealed that there are needs to include other extension innovations with the current programs. Proposing a synergetic cotton extension model including the modern extension innovations considering the changes occurring in the cotton sector is the need of hour. This paper attempts to review and document the role played by the various Cotton Extension agencies to increase the production of cotton in India.

India is one of the important contributors of world cotton production and cotton plays a significant role in Indian economy. Over the years the area, production and productivity of the crop has undergone significant changes. Many factors were responsible for the desirable changes occurred in the production level of cotton in the country *viz.,* the researchers, cutting edge technologies, development agencies, extension programs, policy support and hard working Indian cotton growers. Among them, the efforts taken by the various cotton extension agencies and the effectiveness of the cotton extension programs executed are very vital to document. This paper attempts to review and document the role played by the various cotton extension agencies to increase the production of cotton in India.

Development of Institutions for Catering the R and D needs of Cotton Improvement in the Country : The establishment of various institutions in India in 20th and 21st centuries accelerated the outreach programs of cotton. With the establishment of Agricultural Departments in various provinces of India in 1904, the various problems concerning the cultivation of cotton in India were systematically studied. The Department of Agriculture in Bombay, central provinces, Berar, United Provinces and Madras were pioneers in taking up such research works on cotton. To meet out the demand of Lancashire industry, Indian Cotton Committee was set up in 1917, to investigate the possibilities of extending the long staple cotton in India. Based on the recommendation of the committee, Indian Central Cotton Committee

(ICCC) was set up at Bombay in 1921 as a technical advisory board to Government. In 1924, the ICCI set up Cotton Technological Research Lab under it aegis at Bombay. From 1924-1937, the committee supported the entire expenditure for cotton Research and Development schemes. In 1938, the ICCI was wound up and its functions were transferred to Indian Council of Agricultural Research (ICAR), New Delhi. In 1959, the "Project Intensification of Regional Research on Cotton, Oilseeds and Millets" (PIRCOM) was started at Coimbatore which contributed significant research works in cotton improvement. In 1966, the ICCI was abolished and in 1967, All India Coordinated Cotton Improvement Project was launched at Coimbatore. In 1976, the central Institute for Cotton Research was started at Nagpur under ICAR. In 1976, the Coimbatore centre which was regional station for Indian Agricultural Research Institute has become the regional centre for CICR, Nagpur to cater the researchable needs of southern zone. In 1985, another regional centre of CICR, Nagpur for northern zone was established at Sirsa, Haryana.

Besides, the Directorate of Cotton Development has been established in April 1966 under the administrative control of Ministry of Agriculture, Department of Agriculture & Cooperation, Government of India. It was established after the abolition of ICCC for monitoring the progress of cotton developmental programs & schemes and other related activities on all aspects of cotton. The Directorate has also been assigned with the duties to disseminate the latest production technologies among the farmers of different cotton growing eco-systems in various states through various schemes. This Directorate monitors the progress of Intensive Cotton Development Program (ICDP) under Mini Mission II of Technology Mission on Cotton (TMC) in 13 implementing states like Punjab, Haryana, Rajasthan, Maharashtra, Gujarat, Madhya Pradesh, Karnataka, Tamil Nadu, Andhra Pradesh, Orissa, Uttar Pradesh, West Bengal and Tripura as nodal office. Beside this, this Directorate is also monitoring the progress of other centrally sponsored schemes being implemented in the states of Maharashtra, Gujarat and Goa as area office. In addition, the Cotton Corporation of India, a profitable and MOU signing Company with the Ministry of Textiles, Government of India was established. It is an institute, championing the interest of Indian cotton farmers for more than three decades as a single largest cotton trading company in the country. It operates more than 300 procurement centres to ensure fair price for the stakeholders.

First Line Extension Programs of ICAR and other Cotton Extension Programs Implemented in the Country : Realizing the scope and importance of extension functions, the ICAR established a section of Extension Education at its headquarters in 1971 which was later on strengthened and renamed as Division of Agricultural Extension. It was intended to enforce this functional relationship down the line in the research institutes, agricultural universities and allied institutions. There were four main Transfer of Technology (ToT) Projects of the ICAR, namely National Demonstrations (ND), Operational Research Project (ORP), Krishi VigyanKendra (KVK) and Lab to Land Project (LLP). These first line extension programs of Indian Council of Agricultural Research also facilitated the effective transfer of latest cotton technologies both in last as well as in the current century. To ensure the transfer of technology in cotton crop, along with the first line extension programs of ICAR, several other programs viz., Front Line Demonstrations (FLD), Integrated Pest Management (IPM), Integrated Resistance Management (IRM),

Technology Assessment and Refinement (TAR) - Institute Village Linkage Programme (IVLP), Intensive Cotton Development Programme (ICDP), Farmers Field Schools (FFS), National Agricultural Development Project (NADP), National Agricultural Innovation Project NAIP, Agricultural Technology Information Centre (ATIC) etc., have been launched and are being implemented with the active cooperation of the ICAR Institutes, State Agricultural Universities and Extension personnel of the State Department of Agriculture. In later years, some of the programs were merged with the newly started programs. One such program is the Intensive Cotton Development Programme which was started three decades ago in 1971-1972 was later merged with MM II of Technology Mission on Cotton (TMC) in 2000.

Front Line Demonstrations by AICRP on Cotton : Among the various first line extension programs of ICAR, the FLDs a typical kind of national demonstrations played a major role in disseminating novel cotton production and protection technologies to cotton growers. Since 1996-1997 crop season, the AICCIP has been conducting FLD on cotton through its networking centers and by the CICR and its regional stations in Coimbatore and Sirsa. It has the objectives of demonstrating the usefulness of the latest improved crop production and protection technologies to the farmers as well as extension workers with a view to reduce the time gap between technology generation and its adoption, enabling the scientists to obtain direct feedback from cotton farmers and suitably reorient their research programs and develop appropriate technology packages and to create effective linkage among scientists, extension personnel and farmers. Until 2013, these demonstrations were conducted on Cotton Production Technology, Integrated Pest Management and on Farm implements. High yielding varieties and hybrids suited for various agro climatic conditions approved transgenic cotton hybrids, Integrated Nutrient Management (INM), use of bio fertilizers, bio pesticides, water management, intercropping system, IPM, cotton farm implements etc., were the technologies demonstrated through this type of FLD. So far a total of Seven hundred and fifty seven lakh rupees had been spent by the Ministry of Agriculture for conducting 16827 FLDs on cotton production technology, 148 unit demonstrations of cotton Integrated Pest Management and 125 unit demonstrations of cotton farm implements in the ten cotton growing states of India. Since 2014-2015, the FLDs are being conducted under National Food Security Mission – Commercial crops by Government of India. Under this mission, FLDs are being conducted on Integrated Crop Management, *Desi* / ELS cotton / Seed production and Intercropping in cotton.

Technology Mission on Cotton : Considering the importance of cotton crop in the national economy, Government of India has launched Technology Mission on Cotton (TMC) in February 2000 with the objective of improving production, productivity and quality of cotton so as to enable Indian cotton to compete globally in the free market economy under the WTO regime. Four Mini Missions have been established to fulfill the aforesaid objectives. Mini Mission I with the Indian Council of Agricultural Research to look after the research on cotton, Mini Mission II is with the Ministry of Agriculture, Department of Agriculture and Cooperation to transfer of technology for cotton development and Mini Mission III and IV with Ministry of Textile to develop market infrastructure and modernizing Ginning and Pressing factories respectively. The synergy to increase productivity with lowered farm-level cost of

production is the major aim of the Mini Mission I with emphasis on quality and global competitiveness. Technologies generated out of judicious and logical integration of various location-specific farm-resources are the primary target of this Mini Mission. Mini Mission II, on the other hand, aims at increasing production and productivity of cotton, making available the quality seeds of improved varieties / hybrids to the farmers, transferring production technology to farmers through front line demonstrations and training of farmers/extension workers, bringing more area under irrigation and promoting efficient use of water by popularizing drip and sprinkler irrigation, minimizing losses to cotton crop by pests through popularizing IPM module and IRM strategies, pest surveillance etc., (AFC, 2010)

ICT initiatives in Cotton TOT programs : Viewing the modern advancements in ICT and advantages in mobile phone technology, the Central Institute for Cotton Research functioning under the Indian Council of Agricultural Research has been executing a novel extension mechanism called "e-*Kapas* network" for effective knowledge transfer among Indian cotton growers in the current plan period. "e" meant for electronic and "*Kapas*" in Hindi (one of the major Indian languages) means cotton. 'e - *Kapas*' essentially refers to the utilization of electronic devices - mobile phones for delivering cotton technologies to farmers, extension workers and other development workers engaged in cotton sector. The project is functioning under Technology Mission on Cotton Mini Mission I, to increase the productivity of cotton in the country. The project has been functioning in 17 centres across the ten cotton growing states of the country under the leadership of Central Institute for Cotton Research, Nagpur. Farmers interested in e-Kapas network register with their local state centres by registering their mobile numbers. Centres send regular Voice SMS about cotton genotypes, production and protection technologies in their local languages to the registered growers (Usharani *et al.,* 2014). Under this project, in Tamil Nadu state alone, up to October 2015, 18479 farmers had registered and so far 5,27,430 voice SMS alerts have been sent to the registered growers.

Challenges ahead : Analysis on all these cotton extension programs revealed that they were effective in some aspects viz., in increasing the yields, sharing the knowledge but handicapped due to lack of professional execution and non-availability of latest technological dissemination tools for ready transfer (Usharani and Wasnik, 2011). Many of them excluded the novel extension innovations *viz.,* cyber extension, market led extension, farmer-led extension and environmental extension for a wider reach. Hence, developing a cotton extension program with inclusive extension innovations is a challenge.

Also the major cotton Transfer of Technology efforts tried so far to disseminate the innovations and bridge up the gap viz., Front Line Demonstrations, Farmers Field Schools etc., were basically developed for other crops in other countries and later replicated in cotton. Hence, developing an exclusive Indianized cotton extension program is a challenge.

Also, the Indian cotton sector is facing serious challenges posed by the changes viz., changing technology "*Bt* cotton", changing demands of the textile industries and non woven sectors and changing scenario of attaining top position in acreage and production at world level. Developing an extension program which can cope up the changes in cotton sector is another challenge.

Even though India owns the laurels of being first in world cotton acreage and second in production, it has been facing challenges with regard to increasing and sustaining the crop's productivity for many years. Many technologies released from the cotton research system could not bring out a breakthrough in increasing the productivity. Among the various reasons cited for less productivity, lack of information about available yield enhancing cotton technologies is one among the major ones. The information and communication support for this crop during last 57 years has mainly been conventional. The cotton technologies spread through extension personnel of the state department of agriculture was mostly manual. This approach has not been able to reach majority of the cotton farmers who are spread across the whole country (Usharani *et al.,* 2014). This gap remains a challenge for the cotton extension system even today.

Even though, the introduction of "e-*Kapas*" nullifies the criticism that results and advisories of cotton research did not reach the farmers in time, still there are needs to include other extension innovations and technology forecasting aiming inclusive development with the current program (Usharani, 2015).

Future Perspectives : The challenges posed by the changes in the cotton sector force the extension system to arrive a novel extension approach for future cotton farming in the country. Hence, proposing a synergetic cotton extension model for profitable and sustainable cotton farming with due consideration of changes occurring in the cotton sector is the need of hour.

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Efforts by the Union Ministry of Agriculture and Farmers Welfare through implementation of National Food Security Mission - Commercial crops (NFSM –CC) for improvement in production scenario of cotton

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Cotton is one of the principal commercial crops. It plays an important role in the National economy providing large employment in the Farm, Marketing and Processing sectors, besides providing the basic input for Ginning and Pressing, Textile Industry, Import and Export of yarns fabrics etc. This crop is cultivated across the world by about 80 countries in an average area of 337 lakh hectares (2009-2010 to 2013-2014) receiving 1479 lakh bales (170 kg each) of production with the productivity of 746 kg /ha. India is at first place by contributing 34 per cent in area (113 lakh ha), second place in production by contributing 22 per cent (318 lakh bales) after China. During the same period the average consumption of lint in the world was 1486 lakh bales including 276 lakh bales in India. India is self sufficient, however, the average productivity is 478 kg/ha against the world's average of 746 kg/ha showing a gap of 268 kg/ha (less by 36%). The reasons responsible for low productivity are weather aberrations including inadequate / excess rains with un even distribution of rains in cotton producing areas, more than 65 per cent of area rainfed in central and south zones and more incidence of pests, especially sucking pests in *Bt* cotton etc.

The National Development Council (NDC) in its 53rd meeting held on 29th May, 2007 adopted a resolution to launch a Food Security Mission comprising Rice, Wheat and Pulses to increase the additional production of Rice by 10 million tonnes, Wheat by 8 million tonnes and Pulses by 2 million tonnes by the end of the Eleventh Plan (2011-2012). Accordingly, a Centrally Sponsored Scheme, 'National Food Security Mission' (NFSM), was launched in October 2007. The Mission met with an overwhelming success and achieved the targeted additional production of Rice, Wheat and Pulses. On this basis, the Mission is continued during 12th Five Year Plan (2012 - 2013 to 2016- 2017) with new targets of additional production of food grains of 25 million tonnes comprising of 10 million tonnes of Rice, 8 million tonnes of Wheat, 4 million tonnes of Pulses and 3 million tonnes of Coarse cereals by the end of 12th Five Year Plan (2016- 2017). The ongoing schemes of Initiative for Nutritional Security through Intensive Millets Promotion (INSIMP) Programme, Maize component of Integrated Scheme of Oilseeds, Pulses, Oil palm and Maize (ISOPOM), Dual purpose coarse cereals of Accelerated Fodder Development Programme (AFDP), Technology Mission on Cotton (TMC) and Jute Technology Mission (JTM) discontinued and

merged with National Food Security Mission' (NFSM). Now, there are five components under the ambit of NFSM i.e. NFSM-Rice, NFSM-Wheat, NFSM-Pulses, NFSM-Coarse cereals and NFSM-Commercial Crops (NFSM-Cotton, NFSM – Sugarcane and NFSM- Jute).

Under the National Food Security Mission (NFSM): Commercial Crops, NFSM: Cotton has been approved by the Government of India for implementation in 12th Five Year Plan. The details about NFSM –Cotton are as follows:

i) Name of scheme: National Food Security Mission: Commercial Crops: Cotton (NFSM-CC-Cotton).

ii) Type: Centrally Sponsored Scheme.

iii) Year of commencement: 2014-2015.

iv) Objective: To enhance production and productivity of cotton.

v) Salient features: Components of NFSM-CC-Cotton are Insecticide Resistance Management (IRM), Online Pest Monitoring and Advisory Services (OPMAS), Frontline Demonstrations: 1) FLDs on Integrated Crop Management (ICM), 2) FLDs on Desi and Extra Long Staple (ELS) Cotton/ FLDs on ELS cotton seed production, 3) FLDs on Inter-cropping, Trials on High Density Planting System (HDPS) and Monitoring, evaluation and electronic print media.

vi) Structure of Scheme: NFSM-CC-Cotton thrusts on transfer of technology through demonstrations and training in order to extend benefits to the farmers.

vii) Funding pattern: 1) General Category Cotton Producing States: 60:40 (Sharing pattern between Central and State Governments), North Eastern and Hilly States: 90:10 and Central Agencies 100 per cent.

viii) Eligibility: Project based assistance is provided to farmers / States / implementing agencies.

ix) Area of operation : 15 States: Andhra Pradesh, Assam, Gujarat, Haryana, Karnataka, Madhya Pradesh, Maharashra, Orissa, Punjab, Rajasthan, Tamil Nadu, Telangana, Tripura, Uttar Pradesh and West Bengal.

x) Procedure to apply: Project proposals duly submitted by the State Governments / other agencies are considered for assistance under the scheme as per its guidelines / norms.

xi) Budget allocation in lakh year wise: 1) 2014-2015 : Rs 1200 lakh (100 % funding to all implementing agencies) and 2) 2015-2016: Rs. 1673 lakh (Total outlay of scheme including Central and states share and Central agencies 100%). During 2015-2016, NFSM is a part of **Krishi Unnati Yojana (State Plan).**

4. Area, Production and Yield scenario of cotton under NFSM-CC-Cotton: During the first year of implementation (2014-2015), the total area sown with seeds of cotton was 130.83 lakh hectare including 119.70 lakh hectare under Bt. Cotton, from which, 354.75 lakh bales produced with productivity of 461 kg lint/ha. As per first advance estimate of area, production and yield during 2015-2016, about 335.07 lakh bales of cotton production from 117.63 lakh hectare area is estimated with productivity of 487 kg/ha. The aforementioned figures indicate that area and production of cotton reduced to

States	2014-2015*			2015-2016**		
	A	P	Y	A	P	Y
Punjab	4.20 (4.08)	16.00	648	4.50(4.44)	16.82	635
Haryana	6.47(6.31)	23.00	604	5.86(5.27)	23.0	667
Rajasthan	4.87(3.95)	16.00	559	4.06(3.56)	13.0	544
A. North Zone	**15.54(14.34)**	**55.00**	**602**	**14.42(13.27)**	**52.82**	**623**
Gujarat	30.10(27.13)	110.89	626	27.61(26.23)	108.0	665
Maharashtra	41.92(40.10)	70.19	285	38.24(34.40)	73.41	326
Madhya Pradesh	5.74(5.50)	17.50	518	5.47(4.86)	17.0	528
B. Central Zone	**77.76(72.73)**	**198.58**	**434**	**71.32(65.49)**	**198.41**	**473**
Telangana	17.20(16.08)	38.00	376	16.89(16.61)	47.0	473
Andhra Pradesh	8.20 (8.00)	28.41	589	6.62(6.50)	15.0	410
Karnataka	8.69 (6.97)	22.00	430	5.87(4.87)	11.50	338
Tamil Nadu	1.86 (1.58)	7.86	718	1.05(0.90)	5.34	923
C. South Zone	**35.95(32.63)**	**96.27**	**454**	**30.43(28.88)**	**78.84**	**448**
Odisha	1.27	4.00	535	1.25	4.0	544
Others	0.31	0.90	494	0.21	1.0	810
D. Other than A,B,C	**1.58**	**4.9**	**555**	**1.46**	**5**	**582**
Total (A+B+C+D)	**130.83(119.70)**	**354.75**	**461**	**117.63(107.64**	**335.07**	**487**

*Fourth advance estimate, **First advance estimate, () Figures in parenthesis indicate area under *Bt* cotton in lakh ha, A: Area, P: Production, Y: Yield.

10.08 and 5.55 per cent respectively in 2015-2016 over total area and production of cotton in 2014-2015, but productivity showed a marginal increase of 26 kg/ha in 2015-2016. Reasons for reduction in area in north zone appeared like receipt of rains at grain maturity / harvesting of rabi crops 2014-2015 delayed sowing of cotton and late supply of canal water to some cotton areas in Haryana and Rajasthan in north zone. In Central and South zones, the farmers shifted the cotton area to some other crops like soybean etc due to late receipt of monsoon rains.

A number of reasons are responsible for reduction in cotton production during 2015-2016. But major ones appeared are infestation of whitefly in cotton in north zone, especially in Punjab. Long dryspells and early withdrawal of monsoon made cotton plants stunted, particularly in upland areas of central and south zones. As a result, plants produced less number of bolls, which resulted in reduction of cotton production. The state wise details of area, production and yield during NFSM: CC: Cotton *i.e.* 2014-2015 to 2015-2016 are shown in table below : Area: Lakh ha, Production: Lakh Bales, Productivity: Lint kg/ha

The farmers of cotton producing states are suggested to take advantage of newly technology demonstrated at their farms by the expert scientists / extension personnel of Indian Council of Agricultural Research (ICAR) / State Departments of Agriculture (SDA)under NFSM: CC: Cotton and make all efforts to cultivate cotton scientifically for harvesting improved quality production with accelerated productivity for getting remunerated price for their produce.

Futuristic Extension Model for empowerment and improvement of livelihoods of small scale cotton farmers through Farmer field Schools, Farmer Life Schools and Supply Chain Linkages* - A better cotton initiative

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ABSTRACT : A programme on better cotton initiative was launched by Participatory Rural Development Initiative Society (PRDIS), with support from better cotton fast track fund (BCFT) and other partners during 2010-2011 in selected villages of Mahabubnagar district of Andhra Pradesh, involving 2000 small scale cotton farmers. The programme is scaled up serving 21000 farmers in Andhra Pradesh and Telangana States during 2015.

The programme aims to empower farmers on better cotton production principles through Farmer Field Schools (FFS) and decent work principles through Farmer Life Schools (FLS) as well as supply chain linkages through farmers organizations namely groups networks/Federation and Producer Companies. During this process two producer companies were formed and they are immensely benefitted through collective bargaining on inputs and marketing. Besides the information, communication technologies (ICT) were used for field monitoring and guidance.

The programme has amply demonstrated as an holistic and futuristic agricultural extension model linking production with markets; The potential of FFS and FLS as an innovative extension methods for empowerment and improvement of livelihood of cotton farmers. In addition to improvement there was increase in human, social and economic capital Majority of farmers were able to get net additional income of about Rs 10000 to 15000/ac, 80 per cent reduction in pesticide usage 25 per cent reduction in chemical fertilizers and changes in attitude towards health, hygiene and environment and application of decent work principles specially in not using banned pesticides, child labour and awareness on farmers rights. Thus the programme has shown the power of changing the lives and livelihoods of cotton farmers through a futuristic extension model.

Globally and domestically, cotton is an important agricultural commodity. In India, Cotton exports are not only a source of vital foreign exchange earnings, but also account for a substantial proportion of their GDP and tax income, leading to significant economic and social development. About 70 per cent of the global cotton production comes from 4 countries, which include China (27%), India (22%), USA(13%) and Pakistan(8%).

India is a major producer of cotton ands is also the 2nd largest exporter after the USA. It accounts for around 59 per cent share in the raw material consumption baskets of the Indian textile industry. Thus cotton plays a major role in sustaining the livelihood of an estimated 5.8 million cotton farmers and about 40-50 million people engaged in related activities , such as cotton processing and trade. India has largest cotton cultivated area, which constitutes about 30 per cent of the global cotton area. India's cotton production has been increasing from

2003 to 2001 as a results of a range of initiatives, such as better technology, seeds, nutrients management, irrigation and governmental initiatives etc.

About 70 per cent of total cotton production is contributed by 3 states: Gujarat (26%), Maharashtra (22%) and Andhra Pradesh (23%), where as 70 per cent of cotton is consumed by spinning mills located in Tamil Nadu, Maharashtra, Gujrat and Punjab. Approximately 65 per cent of India's cotton is produced on rainfed areas.

Accelerated demand for cotton, globally, has led to more than threefold increase in its production since 1950s. This increase in production has been achieved through intense inputs application, use of which has most often overlooked environmental impacts. Some of these unsustainable production practices include indiscriminate use of pesticides and fertilizers, extensive use of irrigated water , with no regard to water quality and quality, use of application that contribute to soil erosion, and an unbalanced (quality, time of use) use of resources in some areas.

The environmental impacts associated with cotton production, such as soil and water pollution, are increasingly coming into focus and reiterating the need for sustainable production systems. The united nations defines 'sustainable development 'as development that meets the needs of the presents, without compromising the ability of future generations to meet their own needs. The basic premises of this definition are supported by there pillars of sustainable development economic sustainability, environmental protection and social security.

The better cotton initiative programme (BCI) : Keeping in view of sustainability of cotton, better cotton initiative is global voluntary initiative formed, supported by a range of stakeholders, such as producers, global retailers, traders and financial institutions. The purpose of BCI is to promote measurable improvements in about 21 countries including the key environmental and social impacts of cotton cultivation, by mainstreaming sustainable production practices through implementing partners and its local partners. The programme is being implemented in India, China, Mosanbique, Pakistan, Brazil, USA, Turkey, Kenya, Mali, Uzbiksthen, Austtralia etc., In India the programme is implemented in 11 States including Andhra Pradesh and Telangana

The core components of the Better Cotton System comprise:

- Production principles and criteria to provide a globally acceptable better cotton
- Farmer support to promote enabling mechanisms at a local and global level, working with experienced implementing partner , and stimulating public private partnership funds to implement these mechanisms
- Farm assessment to encourage farmer to continuously improve, through measuring results and seasonal learning cycles
- Supply chain connecting supply with demand through an identifiable bale of 100 per cent better cotton lint
- Monitoring, evaluation and learning mechanisms to measure progress and change and to ensure the better cotton systems has the intended impacts on its direct beneficiaries

In other words the purpose of BCI is to promote strong supply chain linkages with measurable improvements in productivity and reduction in cost of cultivation besides

environmental concerns.

PRDIS initiative : As an endorsed BCI implementing partner, solidaridad manages several projects in India in collaboration of local partners. PRDIS is chosen as local Partner (during 2010-2011) based on its successful track record on productivity enhancement programmes over a decade. PRDIS worked with 2000 farmer (1000 farmers each from Boothpur and Bijinepally mandals with eight 8 villages in each mandal) are grouped into 81 learning groups (LGs) with eight (8) villages in each mandal) are groups were empowered through farmer field schools and farmer life schools. The PRDIS later recognized as on Implementing Partner (IP) and worked in Andhra Pradesh, Telangana, Maharastra and Karnataka States with about 12000 farmers. Which were increased to 21000 farmers during 2015.

a. Farmer Field Schools (FFS) : It is an approach through which farmers undergo a field oriented, discovery based training that enables them to become field experts, be able to grow a healthy crop and promote better quality of life in a healthy environment. FFS is recognized as an effective extension tool, which can be used for empowering the farming community, development self confidences of people and improvement in social, economic and human capital. A unit of FFS generally includes 25 farmers. A facilitators assists this groups in order to builds the capacity of farmers, the facilitators of PRDIS mainly organized Farmer Fields Schools in all villages where in selected members of each LG were enrolled to empower them on knowledge and skills of BCI production principles and minimum production criteria (include crop protection practices, Water , Health of the soil, Natural habitats, Biodiversity, Quality of the fibre etc). In addition , farmer were also taught the mechanics of groups dynamics. The trained farmer in turn empowered the other member of the members of the groups.

Further this method had good impact on farmers since regular field monitoring with cotton Eco system Analysis (CESA), it was Possible to take rational decisions on resource use management. The trained farmers in turn were intern have motivated and guided other farmers in their LGs for implementation of knowledge and cotton eco system analysis, decision making skills on IPM, INM, ICM, soil , water, fibre quality , harvesting etc.

Prior to the launch of this programme, a baseline survey was conducted to know the basic information on cotton cultivation practices including land preparation, seed selection, inter cultivation, crop nutrition, pest management , harvest , and marketing , cost benefit ratio. Identified gaps are the farmers are growing *Bt* cotton with out refugee crop, boarder crop, inter crop, trap crop and imbalanced fertilization and spray banned and dangerous chemical pesticides for sucking pest management.

b. Farmer Life Schools (FLS) : Similarly, the Farmer Life Schools (FLS) an extension of FFS were organized for empowering farmer on decent work principles such as (farmer associations, forced labour child labor, for hazardous work the minimum age is 18 year, Non discrimination, basic treatment and displinary practices, life skills and livelihood skills etc.)

Farmer life school is an open school used for empowerment of farmers to lead a quality, problem free and happy life with self help, mutual helps and cooperation.

All farmer were taught and given hands on experience on problem analysis,

prioritization, solutions to identified problems and implementation arrangements specially on child labor and other issues

c. ICT interventions : In addition to the above method ICT tools were used to supplement and compliment them for find traceability, monitoring, advise and guidance**.**

d. Cotton supply chain in India : The agriculture produce marketing committee (APMC) is the primary market infrastructure in the country through which cotton is marketed. The main function of these markets or mandal is to regulate market practices such as weighing, Process of sale, method of grading, payment process, etc. APMCs also provide facilities for storage, boarding and lodging for buyer , sellers , etc .this committee charges 1 per cent of the goods value as fees from the buyer .the marketing committee , which runs the market and developing the markets yard for and sellers who have the responsibility of maintaining and developing the market yard for its users.

The three Marketing Agencies engaged in cotton trade

- Private sector comprising traders, owners of ginneries operating as individual business proprietors, partnership firms and private limited companies.
- Public sector agencies like the Cotton Corporation of India (CCI)
- Co operative sector

It has been estimated that approximately 80 per cent of the marketed surplus of *kapas* and lint is handled by the private marketing channels and the remaining 20 per cent by the institutional marketing channels including co operative and Cotton Corporation of India (CCI)

In private setup, farmer sells cotton directly to ginners, primarily in the form of kapas (seed cotton). Recently , aggregators have started to play a major role in collecting raw cotton from farm gate of 10-15 farmers and in selling the consolidated produce to ginner in a radius of 100-150km sometimes if the prices are attractive the consolidated cotton is dispatched t over 200km from Maharashtra to Gujarat.

e. Organizing Producer Company for connecting supply with demand as well as sustainability of the project : During the year farmers were given knowledge on association of farmer groups to take up marketing linkages with ginners. Efforts are also made with few ginners situated in the district to get cotton directly from farm gate. This year two Producer Companies were formed involving all stakeholders as members to take up activities for sustainable cotton value chain. The cotton value chain has several links starting from monsoon to market such as plant population Bio diversity input utilization, fibre quality storage and supply chain links

f. Supply chain and traceability

- Cotton from farms is proposed to extract fibre from the seed in a ginning unit and the lint is packed in 170 kg bales
- These bales are loaded and transported to spinning mills to manufacture yarn
- Yarn is a raw material used by weaving units to manufacture grey fabric.
- Grey fabric is dyed and finished for providing color and property to the cloth in process houses.
- Finally, the dyed and finished cloth is used in the garment manufacturing unit to stitich various clothes

- The better cotton can be traceable from level to manufactures by all stakeholders

Monitoring, Capacity Building and Credibility Checks : Periodical monitoring were undertaken by the consultants of PRDIS and Solidaridad in addition to undertaking capacity building programmes like training of trainers by Solidaridad. Training of facilitators by consultants of PRDIS, training of farmers by facilitators using Farmer Field Schools approach for production principles and farmer life schools approach for decent work criteria (Freedom associations, forced labour, child labor, For hazardous work the minimum age is 18 yrs. Non discrimination, Basic treatment and displinary practices etc). Besides, self assessment is done by LG members to encourage farmers to continuously improve, through measuring results and seasonal learning cycles, followed by 2nd arty credibility check is done by a team comprising of members from Solidaridad and PRDIS. Finally 3rd party verification id done by an agency appointed by BCI in order to issue a license to farmers in addition to connecting supply with demand through and identifiable bale of 100 per cent better cotton linit.

This year PRDIS was choosen by BCI in India for impact assessment study in allian with iseal. Baseline survey has been successfully completed.

Achievements and Benefits to the Farming Community

Improvement in Human Capacity through various capacity building programme : Farmers were empowered on knowledge (90%) and skills (80%) concerning IPM practices, banned pesticides (100%), labeled and graded pesticides (100%) water management (90%), soil health management (85%), fibre quality and harvesting as well as post harvest technologies. In addition awareness and attitudinal changes on decent work principles such as use of child labour, discrimination, forced labour, health and safety environment and biodiversity line also emphasized.

Improvement in yield quality and Economics : The analysis of the yield data revealed that was incremental yield of 25-30 per cent derived out of BCI efforts by about 80 percent of farmers. The cost of cultivation was also reduced by about Rs. 4000 / 6000/ac (reduction use of pesticides 80%) fertilizers 25%). The net additional income gained by BCI farmer compared to control group was Rs.10,900 to 15,000/ac. The quality of produce was also without contamination and excess moisture. On the other hand, the programme has demonstrated the production potentiality through FFS which has recorded 1.7 q/hector. This gap PRDIS will bridge during the coming years. Besides, the cost of cultivation also will be further reduced through soil test based fertilizer application and agro eco system based resource management. This will provide more net income to BCI farmers.

Improvement in Social Capital : The farmers were sensitized about the need for forming the groups and associations (producer company) for collective bargaining, to demand for their rights and directly linking the supply wit demand.

Other benefits

- All the BCI farmers have not used the child labour

- Majority has not used forced labour and children below 18 years for pesticide spray
- There were no discrimination in payment of wages for the same type of work between men and women
- Bio diversity, health, safety, and environmental concerns were taken care off. In fact non pesticide management with use of botanical and bio agents were encouraged.

Leading the change – Conclusion and Lessons : This programme has amply demonstrated (the holistic development) the potential of Farmer Field Schools and Family Life Schools as the innovative extension methods for sustainable holistic development and empowerment of cotton growing farmers. Through the BCI, to farmers human, social and economic capital has considerably improved in terms of knowledge and skills of Better cotton practices and decent work. Majority of farmers were also able to get net additional income of about Rs 10,000/ha, reduced pesticide usage changed attitudes on environment, health and safety concerns as well as awareness on to need to organize producer organization / company for collective bargaining of fair prices of their commodities. Thus the programme has shown the power of changing the lives and livelihood farmers in the project area directly and about equal number of farmers indirectly. This futuristic agricultural extension model can also be extended to different crops and enterprises for improving the livelihoods and sustainable agricultural development of millions of resource poor small scale farmers of India.

Farmer Field School (FFS) is an effective extension tool to kindle the hope and to meet the educational needs of farmers. This powerful tool has a promise and potential for creating a quite revolution in agricultural and rural development thorough empowerment of farmers

- Prof.S.V.Reddy

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Bt cotton and arthropod biodiversity

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Abstract : Cotton is an important commercial crop in India and plays a key role in national economy. Insect resistant *Bt* cotton has been rapidly adopted during the past twelve years and the commercial approval of *Bt* cotton in 2002 was a breakthrough step to revive the ailing cotton sector in the country. Coincidental with the steep increase in adoption of Bt cotton between 2002 and 2014, the average yield of cotton in India, which used to have one of the lowest yields in the world, increased from 308 kg/ha in 2001-2002, to 541 kg/ha in 20014. The cotton farmers, both large and small holders benefitted from this technology through reduced cost of cultivation, reduced the numbers and volume of insecticide sprays, bollworms population, production cost and environmental contamination, convenience of crop management and increased productivity. However, reduction in insecticide sprays, especially during flowering and boll formation has led to resurgence of some minor pests such as tobacco caterpillar, mealy bugs, thrips, aphids, leafhoppers, green stink bug, and serpentine leaf miner. The reduced insecticide use and personal experience of farmers of not having negative impact of *Bt* cotton on the population buildup of bee colonies, has encouraged beekeepers to keep their bee hives in *Bt* cotton in Haryana, Rajasthan and Punjab in India. Transgenic cotton can have a number of direct and indirect effects on arthropod communities in agro ecosystems. The direct impact is the mortality of bollworms feeding on *Bt* cotton, which can also provide effective or partial control of some other lepidopteran pests and indirectly can affect natural enemies through the removal of eggs, larvae, and pupae of lepidopteran insects that serve as food sources for parasitic and predatory arthropods. There are no adverse effects of *Bt* cotton on the arthropod diversity and natural enemy functions in the cotton ecosystem, and if any are much lower than that of use of insecticides, which have little ecological impact. To prolong the usefulness of the transgenics, important to follow the bio-safety regulations, better presentation of the benefits of biotechnology to the general public, and develop stringent risk assessment and risk management strategies for the safe and rational deployment of transgenic crops for pest management and sustainable crop production.

Cotton is an important commercial crop in India and plays a key role in national economy. India is the first largest global cotton producer and grown in an area of 11.6 million hectares approximately, with about 39 million bales in 2013. For the past two decades, the national average (541 kg/ha) has not reached the world average (785 kg/ha) (Choudhary and Gaur, 2015) creating concern among stake holders. Such a low yield is far from satisfactory and there is scope for further improvement. Of the several factors contributing for low productivity, biotic constraints appear to be very important of which the insects are most important. The pest spectrum includes sap feeders, bollworms and defoliators. Among the pests, bollworm complex is very serious throughout the country and pose a serious threat to cotton cultivation in many agro ecological zones. Cultivation of pest susceptible high yielding varieties and hybrids, resistance in insects to commonly available insecticides, replacement of minor pests into major pests and the phenomenon of resurgence of pests continue to threat cotton production.

Pest scenario : The cotton plant is

infested by pests and diseases right from germination to harvest. More than 160 species of insects infest the cotton plant of which about 10 to 12 species are important depending upon the season and agro ecological situation. The *hirsutum* cotton has replaced the indigenous cotton varieties of *Gossypium herbaceum* Linn. and *Gossypium arboretum* Linn. both in irrigated and rainfed areas. These native cottons were resistant to key pests like leafhopper and bollworm complex. In addition the natural enemy diversity was greater because of a favourable host plant natural enemy interaction on these varieties and the pest were kept under check. The introduction of high yielding varieties of *G. hirsutum* and *G. barbadense* as well as hybrids have altered the pest scenario leading to changes in the composition of herbivores and carnivores and many minor pests have become major ones.

The leafhopper, *Amraaca devastans* (Dist.), whitefly *Bemisia tabaci* Genn. and the bollworm, *Helicoverpa armigera* (Hubner) have attained the status of key pests after introduction of varieties and hybrids. In Punjab, *H. armigera* is reported to complete seven generations in recent years as compared to lesser number of generations in earlier years. Similarly, the cultivation of *G. barbadense* varieties resulted in outbreaks of *Spodoptera litura* Fab. in 1970s and 1980s. Spotted bollworms, *Earias* spp. are serious on *arboreum* than on *hirsutums* in northern states. Staggered sowings, overlapping seasons and indiscriminate use of chemical insecticides resulted in outbreaks of *H. armigera* and *B. tabaci* in peninsular India.

***Bt* cotton in India :** *Bt* cotton is a product of transgenic technology. *Bt* cotton contains the gene *Cry1Ac and Cry2Ab* which provides resistance to the green bollworm, *H. armigera*, the spotted bollworm, *Earias vitella* Fab. and the pink bollworm, *Pectinophora gossypiella* Saunders, all of which are major pests in India. In India, cotton is the first crop to be grown with transgenic technology to manage the bollworm *Helicoverpa armigera*, a serious pest of the crop. The Genetic Engineering Approval Committee (GEAC) of the Ministry of Environment and Forests, Government of India on 26 March 2002 approved three *Bt* cotton hybrids for commercial cultivation in central and southern parts of India. The estimated area under *Bt* cotton in India increased from 600,000 hectares to a record 11.6 million hectares during 2002 to 2014 and production increased from 13 million bales to 39 million bales, respectively. World cotton production was estimated at 151 million bales in 2014, and impressively, India contributed one quarter of this global total. As a result of this phenomenal increase in cotton production in the recent years, India becomes the largest cotton producing country in the world.

In 2014, GEAC's Standing Committee Approved *Bt* Cotton Events and released an additional 70 *Bt* cotton hybrids for a total of around 1167 *Bt* cotton hybrids: the crosses made during the period 2002 to 2014 are predominantly *G. hirsutum* x *G. hirsutum* with a few consisting of *G. hirsutum* x *G. barbadense*. Importantly, India has already achieved a near phasing-out of the Bollgard™1 event, which has now been almost completely replaced with the dual gene Bollgard™ II (BG II) cotton event. During 2014, around ~54 million small holder cotton farmers benefited from planting *Bt* cotton over 11.6 million hectares equivalent to 95 per cent of 12.25 million cotton area(Choudhary and Gaur, 2015).

Benefits from transgenic crops : The large scale adoption of *Bt* cotton in many countries underlines an area wide suppression of target pests that greatly reduces overall

regional populations. Cultivation of *Bt* cotton has resulted in apparent benefits in terms of significant reduction in numbers and volume of insecticide sprays, production cost, environmental contamination, and increased crop yields (Qaim and Zilberman, 2003). The introduction of *Bt* cotton has decreased insecticide use thereby leading to about 4% reduction in the Environmental Impact Quotient (EIQ) in India as compared to 22, 23 and 28% in Australia, USA and China, respectively (Naranjo, 2009). It is estimated that cotton accounted for about 22.5 per cent of total insecticide use worldwide (Anonymous, 1995). However, 45 per cent of the total pesticide consumed was on cotton alone prior to *Bt* cotton introduction in India. With the use of *Bt* technology in cotton, insecticide cost in India has been reduced by 41 per cent. Large reductions in insecticide use have also been observed in other developing countries that have adopted *Bt* cotton (Dhillon *et al.*, 2011). Theoretically, *Bt* transgenic crops limit the exposure of natural enemies to the *Bt* toxins, and reducing the accidental exposure, as typically occurs with conventional insecticides.

Risks : Deployment of insect resistant transgenic plants has raised some concerns about the real or conjectural effects of *Bt* technology because of lack of adequate information. There might be putative risks such as outcrossing through pollen drift, horizontal transfer of transgenes to other organisms, food safety, loss of susceptibility to *Bt* toxins in target pests, disruption of ecosystem processes, direct or indirect effects on nontarget organisms and biodiversity in the tropics. Also the monitoring of genetically modified plants has to be appropriate to detect direct and indirect, immediate and long- term as well as unforeseen effects. Adverse effects of *Bt* toxins on the ladybirds on ingestion of *Bt* fed aphids are unlikely, however, direct exposure to *Bt* toxins or predation on young bollworm larvae on *Bt* cotton plants might have some adverse effects on the ladybirds (Dhillon *et al.*, 2011). At the same time, there is potential for the development of resistance to *Bt* toxins in populations of the target pests leading to failure of this control tactics. Other challenges are manifested through the wide diversity of pests affecting cotton worldwide. For example, *Bt* cotton may indirectly cause existing primary pests or secondary pest problems to increase.

Effect on insect pest complex : Several pest species that are not susceptible to the *Bt* toxins expressed in transgenic cottons also affect cotton production in India (Table 1). There is no evidence of increased susceptibility of *Bt* cottons to nontarget insects such as leafhoppers, red cotton bugs, dusky cotton bugs, green bugs and ash weevils. However, reduction in insecticide sprays especially during vegetative stage has led to resurgence of some minor pests such as tobacco caterpillar (*Spodoptera litura),* mealybugs *(Phenacoccus solenopsis,* and *Maconellicoccus hirsutus),* thrips *(Thrips tabaci)*, aphids *(Aphis gossypii)*, leafhoppers *(Amrasca biguttula biguttula)*, green stink bug *(Nezara viridula)*; and serpentine leaf miner *(Liriomyza trifolii)* in India (Dhillon and Sharma, 2010). Large scale field studies have also confirmed the negative effects of broad-spectrum insecticides on insect communities in both *Bt* and non *Bt* crops (Dhillon and Sharma, 2010 and Naranjo, 2009). Emergence of Mirid bug, *Creontiodes biseratense* has been recorded in its severity on *Bt* cotton in Haveri, Karnataka during 2007. These bugs feed on squares and small developing bolls thereby causing direct damage by yellowing, followed by

Table 1. Major insect pests of Bt cotton in India

Common name	Species	Remarks
Insect pest species which are susceptible to *Bt* cotton		
Bollworm	*Helicoverpa armigera*	Effectively controlled by Bollgard I and Bollgard II
Spotted bollworm	*Earias vitella* and *E. insulana*	Effectively controlled by Bollgard I and Bollgard II
Insect pest species which are not susceptible to *Bt* cotton		
Leafhopper	*Amrasca biguttula biguttula*	Major pest of cotton with increased incidence across India
Aphid	*Aphis gossypii*	Emerging pest of *Bt* cotton in south central India
Whitefly	*Bemisia tabaci*	Major pest across cotton growing regions of India; with outbreaks in 2010 in north and south central India
Red cotton bug	*Dysdercus spp.*	Major pest of cotton with increased incidence
Green stinkbug	*Nezara viridula*	Emerging pest of *Bt* cotton in south central India
Dusky cotton bug	*Oxycaraenus leatus*	Major pest of cotton with increased incidence
Mealy bugs	*Phenacoccus solenopsis and Maconellicoccus hirsutus*	Emerged as major pest of cotton including *Bt* cotton since 2006
Mirid bugs	*Creontiodes biseratense* [Megacoelum=*Lygus biseretensis*]	Recorded as severe pest of *Bt* cotton in Karnataka during 2007
Stem weevil	*Alcidodes affaber*	Serious pest of cotton in southern India since 2003
Grey weevil	*Myllocerus spp.*	Major foliage pest of cotton since 2002
Stem weevil	*Pempherulus affinis*	Major pest of cotton in southern India since 2006
Blister beetle	*Zonabris pustulata*	Emerging pest on *Bt* cotton
*Tobacco caterpillar	*Spodoptera litura*	Emerging as major foliage pest of cotton since 2007
*Pink bollworm	*Pectinophora gossypiella*	Resistance to cry1Ac *Bt* cotton reported in 2009
Serpentine leaf miner	*Liriomyza trifolii*	Major seedling pest of *Bt* cotton in south central India since 2006
Thrips	*Thrips spp*	Emerging as major foliage pest of cotton since 2005

Source: Gujar *et al.*, (2010). *Effectively controlled by Bollgard II.

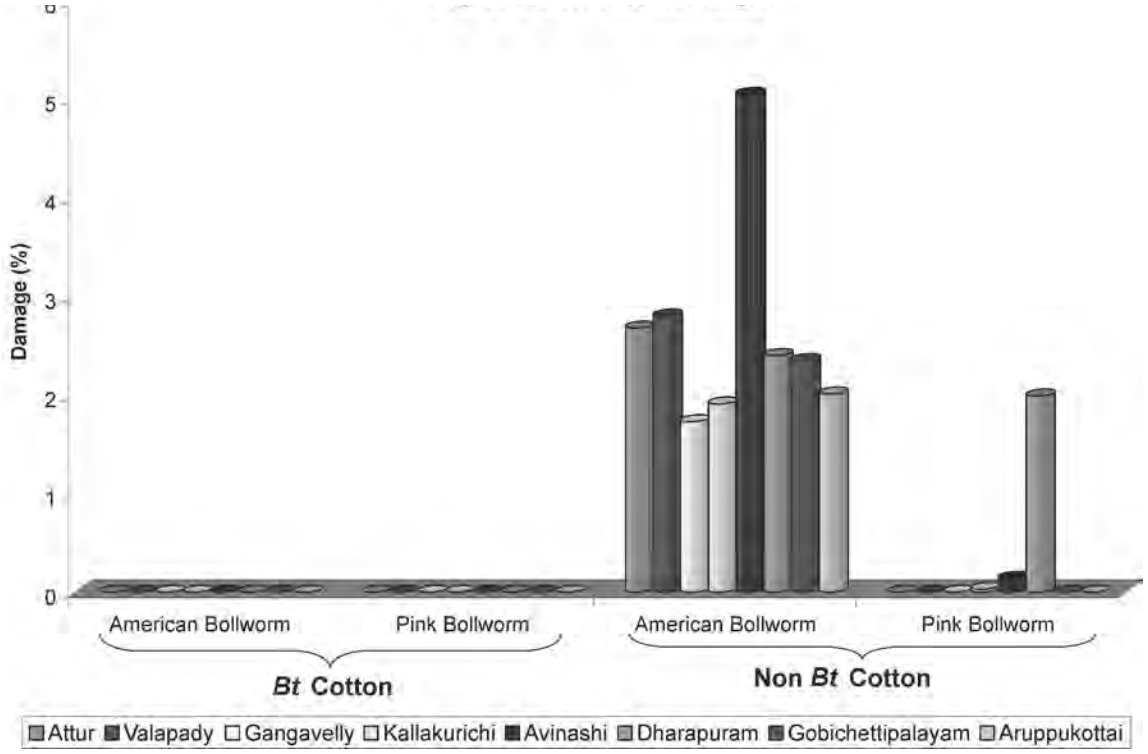

Fig 1. Bollworm damage in *Bt* and non *B*t cotton in Tamil Nadu

shriveling, sharp beak-like twisting, and premature dropping (Patil *et al.*, 2006). Similarly, in China, the mirid bugs (*Adelphocoris* spp and *Lygus* spp) have become key pests of *Bt* cotton.

Effects on arthropod diversity : A number of studies on the influence of transgenics on pests and natural enemies have been carried out to assess their occurrence and ecological consequences in agro ecosystems. Transgenic cotton can have a number of direct and indirect effects on arthropod communities in agro ecosystems. Total arthropods and pests were more abundant in the non-transgenic plots than in the *Bt* cotton plots. The direct impact is the mortality of bollworms feeding on *Bt* cotton, which can also provide effective or partial control of some other lepidopteran pests and indirectly can affect natural enemies through the removal of eggs, larvae, and pupae of lepidopteran insects that serve as food sources for parasitic and predatory arthropods. Men *et al.*, (2003) suggest that the use of transgenic *Bt* cotton decreased the species richness of communities and sub communities. As a result, species composition was altered by eliminating some species and increasing diversity of pest sub communities; however, the diversity of natural enemies was decreased in *Bt* cotton.

Kannan and Uthamasamy (2004) observed significant differences in abundance and species richness between non transgenic and *Bt* cotton. The total arthropods and pests were more abundant in the non transgenic cotton plots than *Bt* cotton plots. *Bt* cotton effectively reduced number of lepidopteran pests such as *Helicoverpa armigera, Earias* sp., and *Pectinophora gossypiella.* However, higher abundance of sucking pests *viz., A. devastans, B. tabaci, A. gossypii* and *S. dorsalis* were recorded in *Bt* cotton than non *Bt* cotton (Kannan *et al.,* 2004 and Jayaprakash *et al.*, 2015). Vennila

et al., (2004) observed varied incidence of sucking pests and bollworms on *Bt* transgenics and non transgenic cotton hybrids; this finding suggests that diversity of arthropods in transgenic crops depends on the host plant and season. On the contrary, Naranjo (2002) reported that natural enemy abundance and overall arthropod diversity are not directly affected by transgenic *Bt* cotton in comparison with non *Bt* cottons.

Long term field studies conducted to compare foliage-dwelling arthropod populations in *Bt* and non *Bt* cotton in south Carolina, Georgia, Northern Alabama, and Southern Alabama revealed that there were no significant differences in the population of arthropod taxa collected stink bugs, plant bugs, *Geocoris* spp., *Orius* spp., *Solenopsis invicta*, ladybeetles, and spiders, while the numbers of *Helicoverpa zea* were significantly lower in the *Bt* cotton fields (Head *et al.*, 2005). Field study on the long-term impact of *Bt* cotton on 22 taxa of foliar dwelling arthropods in Arizona revealed that two taxa declined significantly in unsprayed *Bt* compared with non *Bt* cotton. However, insecticide applications for caterpillars and other pests in both non *Bt* and *Bt* cotton had much greater negative effects on 10 taxa. This long-term study indicated that the effects of *Bt* cotton on non target community are minor, especially in comparison with the alternative use of broad-spectrum insecticides (Naranjo, 2005). Other studies have also reported higher number of arthropods in *Bt* cotton fields under reduced or no insecticide application than in the conventional insecticide protected cotton. Sisterson *et al.,* (2004) did not find significant differences in arthropod diversity in *Bt* and non *Bt* cotton fields. It is expected that in mixed cropping systems that characterizes Indian agriculture, the arthropod biodiversity will either little affected, or if affected, recovers soon. Although, *Bt* toxin have been detected in some

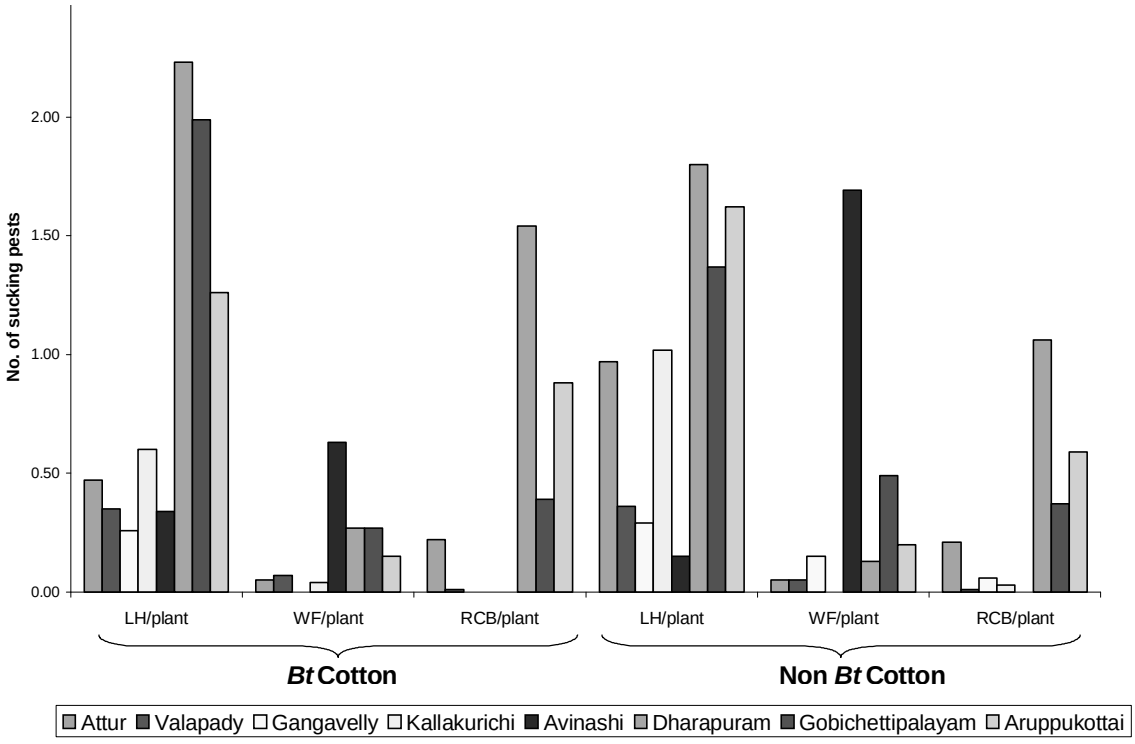

Fig. 2. Incidence of sucking pests in *Bt* and non *B*t cotton in Tamil Nadu

Table 2. Pollinators fauna of *Bt* and non *Bt* cotton hybrids

S.No.	Pollinators	Systematic Position	Relative abundance (%)
1.	*Apis dorsata* Fabricius	Hymenoptera : Apidae	29.83
2.	*Apis cerana* Fabricius	Hymenoptera : Apidae	25.21
3.	*Apis florae* Fabricius	Hymenoptera : Apidae	22.54
4.	*Xylocopa vericalis* Lapel	Hymenoptera : Apidae	
5.	*Ceratina* sp	Hymenoptera : Apidae	
6.	*Thyreus* sp	Hymenoptera : Apidae	
7.	*Megachile lanata* Fabricius	Hymenoptera : Megachilidae	
8.	*Megachile* sp	Hymenoptera : Megachilidae	
9.	*Vespa* sp	Hymenoptera : Vespidae	
10.	*Hypolimnas missipus* Linnaeus	Lepidoptera : Nymphalidae	
11.	*Byblia ilithyia* Drury	Lepidoptera : Nymphalidae	22.42
12.	*Melanitis leda* Linnaeus	Lepidoptera : Nymphalidae	
13.	*Ariadane merione* Cramer	Lepidoptera : Nymphalidae	
14.	*Catopsilia pyranthe* Linnaeus	Lepidoptera : Pieridae	
15.	*Danaus chrysippus* Linnaeus	Lepidoptera : Danaidae	
16.	*Amata* sp	Lepidoptera : Arctiidae	
17.	*Acherontia styx* Westwood	Lepidoptera : Noctuidae	
18.	*Eristalis* sp	Diptera : Syrphidae	

arthropod species, long-term field studies in India on impact of *Bt* cotton on the arthropod diversity indicated that the species richness and diversity index of the plant inhabiting and soil dwelling arthropod species was similar in *Bt* and non-*Bt* cottons. However, insecticide applications in both *Bt* and non *Bt* cottons negatively influenced the arthropod biodiversity of cotton ecosystem.

Bal and Dhawan (2009a and 2009b) reported about 134 species of arthropods, including 54 species of herbivores, 44 species of natural enemies, 26 species of casual visitors and 10 species of pollinators were found to be associated with *Bt* cotton crop. They also stated that among the 134 species, Hymenoptera were most abundant (23.9%) followed by Hemiptera (19.4%), Coleoptera (16.4%), Lepidoptera (14.2%), Orthoptera (8.2%), Diptera and Spiders (4.5% each), Odonata (2.9%), Dictyoptera, Isoptera and mites (1.5%), Neuroptera and Thysanoptera (0.7%) were recorded in *Bt* cotton RCH 134. Kannan *et al.,* (2011) reported that the less incidence of insect pests and higher occurrence of natural enemies were in *Bt* cotton than non *Bt* cotton (Fig. 1, 2 and 3). Dhillon and Sharma (2013) also documented that the total number of insect arthropods on *Bt* and non *Bt* cotton were similar. Also, the Simpsons index of diversity of minor insect species was unity, except for homopterans and suggested that there were no adverse effect of *Bt* cotton on the arthropod diversity under field conditions.

Effects on natural enemies : An important potential effect of transgenic plants is the consequences of changing the occurrence and density of prey for natural enemies. Hilbeck *et al.,* (1998) revealed that green lacewings fed European corn borer (ECB) that had eaten *Bt* corn had a higher death rate and delayed development compared with lacewings fed ECB that had eaten non *Bt* corn. More than 60 per cent of the lacewings fed *Bt* corn reared ECB died compared

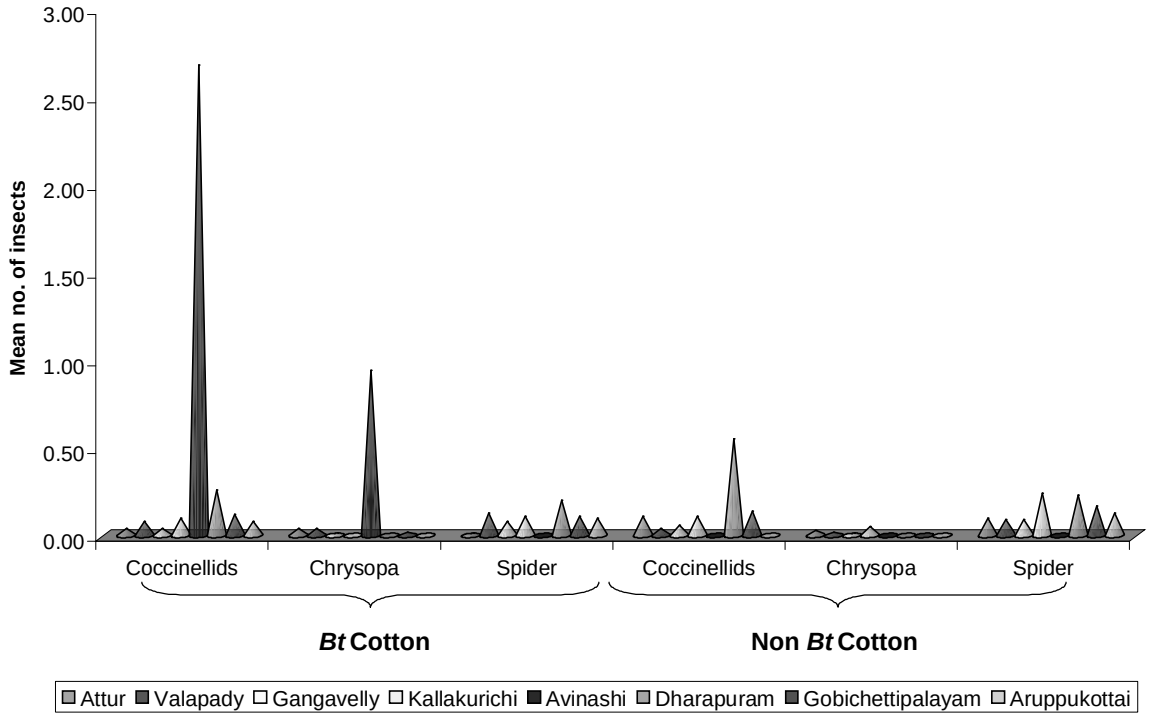

Fig. 3. Population of natural enemies in *Bt* and non *B*t cotton of Tamil Nadu

with fewer than 40 per cent of those fed non *Bt* corn reared ECB. However, when lacewings were fed with spider mites containing the *Bt* toxin negative effects were not observed. Only when lacewings were fed 'sick' lepidopteran larvae reared on *Bt* maize, an effect on green lacewing larvae was noted. However, the possibility in the field that lacewings feed only on lepidopteran larvae would never occur (Hilbeck *et al.,* 1999). Considerable reduction in occurrence of natural enemies population *viz.,* coccinellids, chrysopids and spiders were observed in transgenic *Bt* than non transgenic *Bt* cotton (Kannan and Uthamasamy, 2004). This variation on sucking pests and natural enemies abundance may be due to the interplay of various factors including the abiotic ones.

Naranjo (2002) suggested that natural enemy function such as rate of predation and parasitism of two key pests (*Pectinophora gossypiella* and *Bemisia tabaci*) are unaffected in *Bt* cotton. Predator: prey ratios for *Bemisia tabaci* and *Lygus hesperus* were similar in unsprayed *Bt* and non *Bt* cotton. However, insecticide applications in positive control plots inconsistently altered ratios for *B. tabaci*. Predation indices based on the known feeding activity of selected predators showed that potential predator impact was unaltered by *Bt* cotton but was consistently depressed with insecticide applications (Naranjo, 2005). Whitehouse *et al.,* (2005) reported that the diversity or species richness of the insecticide sprayed cottons in Australia. Monitoring of canopy and ground dwelling predators in commercial *Bt* and non *Bt* cotton fields in Georgia under need based application of insecticides indicated that canopy dwelling predators were not affected by cotton type, while the shift in abundance of some canopy-dwelling

predators was likely associated with insecticide use. These studies concluded that the abundance of predators in cotton fields with standard grower practices failed to exhibit any negative impact of *Bt* cotton on predator populations (Torres *et al.*, 2006). No significant differences have yet been reported in the numbers of generalist predators such as coccinellids, chrysopids and spiders on *Bt* transgenic and their counterpart non *Bt* cottons (Dhillon and Sharma, 2007). Hence the *Bt* transgenic technology does not pose any new threat to natural enemies compared to current pest control practices. Engineering terpene emission to make crop plants favorable to natural enemies is also in progress. Transgenic plants will have to be compatible with natural enemy diversity and dynamism. This being a new dimension offers scope for further study.

Effects on pollinators : Bees and wasps are good pollinators of cultivated crops and can be agents of pollen spread. Bees and bumble bees have been reported to be affected by transgenic products and their systematic study needs to be incorporated into the environmental risk assessment of transgenic plants to make sure that this essential ecosystem service is not damaged (Lovei *et al.*, 2001). Although the toxins expressed in *Bt* corn pollen are specific for Lepidoptera, several studies raise questions about *Bt* effects on pollinators, that is, domesticated and wild bees. The visit and abundance of insect pollinator's *viz., Apis mellifera*, *A. cerana*, *A. dorsata*, *Xylocopa* sp, *Megachile* sp, *Papilio demoleus*, *Hemaris* sp, *Telicota* sp and *Catopsile pyrantha* are higher in *Bt* cotton compared to no *Bt* cotton (Viraktamath and Nachappa, 2004). Presence of pollinating species on cultivated cotton flowers raise the issue of pollen dispersal by insects between *Bt* and non *Bt* cotton, and impact of *Bt* cotton on pollinator species. Studies on prevalence of honeybees (*Apis mellifera*), various Nitidulidae, and Meloidae species (*Mylabris oculata*) on Bt and non *Bt* cotton plants, suggested no impact of *Bt* cotton on their abundance and diversity (Hofs *et al.*, 2008). Vinayakpise and Viraktamath (2015) documented 18 species of pollinators were visiting *Bt* and non *Bt* cotton blossoms (Table 2). Among these, nine species belonged to the order Hymenoptera, eight to Lepidoptera and one to Diptera. These pollinators were common in both *Bt* and non *Bt* cotton hybrids. Among Hymenoptera, *A. dorsata* formed the most dominant pollinator with a relative abundance of 29.83 per cent followed by *A. cerana* (25.21%) and *A. florea* (22.54%). The remaining pollinators constituted 22.42 per cent. As a result of reduced insecticide use in *Bt* cotton in India, beekeepers are keeping their bee hives (*Apis mellifera*) in *Bt* cotton in Sirsa and Dabawali in Haryana, and Sriganaganagar in Rajasthan. These beekeepers are producing good volume of honey, fetching good market price of their honey, and have not reported any negative impact of *Bt* cotton on the population buildup of their bee colonies.

Effects on soil organisms and decomposers : *Bt* toxins can be incorporated into the soil through leaf materials, when farmers incorporate crop residues after harvest. Toxins may persist for 2-3 months, resisting degradation by binding to clay and humic acid soil particles while maintaining toxin activity. Such active *Bt* toxins end up and accumulate in the soil and water from transgenic leaf litter may have negative impacts on soil and aquatic invertebrates and nutrient cycling processes. The toxins, free or bound, had no effect on growth of bacteria, fungi, and algae. Perturbations have

been recorded by several authors with the introduction in the soil of genetically modified micro organisms (such as *Pseudomonas fluorescens* Migula), including displacement of indigenous populations, suppression of fungal populations, reduced protozoa populations, altered soil enzymatic activity, and increased carbon turnover (Stotzky, 2000).

Conclusion : The use of insect resistant transgenic plants will continue to expand and gene pyramiding might become very common in future. However, concerns have been raised about the possibility that large-scale deployment of transgenic crops for pest management might impact the arthropod diversity, natural enemies, toxin flow in the insect fauna through different trophic levels, development of resistance in target insect pests, pollen flow in closely related wild relatives, antibiotic resistance, etc. Similarly, the impacts of transgenic crops on people, on animals, and on the environment are difficult to predict; so it is important that the potential risks be evaluated before transgenic crops are approved for release. The evaluation process inevitably will have to include carefully controlled field testing, bio-safety regulations, to generate information needed to determine to performance of transgenic crops in the farmers field. Refugia can play an important role in resistance management and should take into account the pest complex, the insect hosts and the environment. To prolong the usefulness of the transgenics, the management strategies should reflect the pest biology, insect–plant interactions, their effect on the natural enemies and impact on biological diversity for pest management and sustainable crop production.

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Secondary pests' outbreak in *Bt* cotton

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There are an estimated 67000 pest species that damage agricultural crops, of which approximately 9000 species are insects and mites (Ross and Lembi, 1985). Insect-pests are the major cause of crop losses (Kumar *et al.,* 2008). Averages of 15 per cent of crops worldwide are currently lost to insects (Maxmen, 2013). More than 1326 species of insects have been reported in commercial cotton fields worldwide (Hargreaves, 1948). India is currently the biggest producer of *Bt* cotton in the world since 2012 (Krishna and Qaim, 2012) having largest area under cotton cultivation. Certain pest species of cotton have few deleterious effects on production. These species are called 'minor' or 'secondary' species. The status and the relative economic importance of these different pests vary depending on the agro-ecosystem considered and changes in response to selection pressure to which they are subject.

Commercialized *Bt* cotton through expression of toxic crystalline (Cry) proteins from the soil bacteria *Bacillus thuringiensis* (*Bt*), have effectively managed their target pests (Carriere *et al.,* 2010; Tabashnik *et al.,* 2008). Besides its efficacy against target pest and worldwide adoption, the technology remains under controversy and surrounded by uncertainty (Andow *et al.,* 2009; Lovei *et al.* 2009; Shelton *et al.,* 2009) with respect to potential long term impact especially the development of insect resistance and the impact on secondary insect pests (Lovei *et al.,* 2009; Smale *et al.,* 2006, Garcia and Altieri, 2005).

The Bt cotton has very specific site to act so any detrimental impact on non target organisms (NTO) is expected to be lower than that imposed by broad-spectrum insecticides (Areal and Riesgo, 2015; Guoping Li *et al.,* 2010; Marvier *et al.,*2007; Cattaneo *et al.,* 2006; Naranjo., 2005) applied to manage the primary/ target pests. The reduced use of insecticides may further allow for a higher diversity and density of beneficial arthropods (Lu *et al.,* 2012; Naranjo, 2005). In the absence of target pests, a further concern is that other insect species that are not susceptible to the expressed toxin will develop into secondary pests and cause significant damage to the crop (Wu and Guo.,2005; Sharma and Ortiz., 2000). The development of secondary pests certainly affects other levels of trophic chain which could be of high economic and ecological relevance. The development and effects of secondary pests on *Bt* crops, although of high importance, has received limited attention (Harper, 1991, p.22) and ignoring secondary pests can lead to devastating crop damage that may continue over a considerable period of time. In the event of a secondary pest outbreak, additional pest management interventions are required. The secondary pest's outbreaks and its main causes in *Bt* cotton in India and worldwide has been discussed in the chapter.

Secondary pests in *Bt* cotton crop : Emergence of secondary pests in cotton is a matter of concern as in agricultural ecosystem two relevant phenomena *i.e.* pest resurgence and outbreaks of secondary pests are considered as

ecological backlash events. Where pest resurgence refers to a situation in which a suppressed target pest population unexpectedly rebounds following a pest control action, exceeding the economic injury level (Hardin *et al.,* 1995). But the outbreak of secondary pest is the emergence of a pest other than that originally targeted by an agricultural intervention (the 'targeted' or 'primary' pest), and can be seen as 'replacement' for the primary pest (Hardin *et al.,* 1995; Metcalf, 1980).

A secondary pest is a 'non-targeted pest that has historically posed small or negligible economic threat, but which could be affected directly by a dose expressed in a *Bt* crop, or indirectly through changes in insecticide-use patterns (FIFRA Scientific Advisory Panel ,1998) . Outbreaks of secondary pests is 'an explosive increase in the abundance of a particular species that occurs over a relatively short period of time (Berryman, 1987, p.3) but Metcalf (1986) termed it as 'type II resurgence' which can arise when the primary pest is strongly affected by a pest management strategy, yet is replaced by another pest not affected by this strategy. The causes responsible for both pest resurgence and outbreaks of secondary pests are relatively similar which includes reduction in the number of natural enemies and removal of competitors (Hardin *et al.,* 1995; Ripper, 1956). In broader context, concept of secondary pests is linked with all living organisms that are not affected by newly expressed compounds in *Bt* crops, and can be potentially exposed, directly or indirectly, to the target crop and/or its products in the agro-ecosystem where *Bt* crops will be released or in adjacent habitats' (Arpaia, 2010, p.14). A lethal or sub lethal effect of a *Bt* crop might occur in non target organisms through direct exposure to the *Bt* toxin or indirectly due to changes in the ecosystem on which that species depends (Snow, *et al.,* 2005). To assess the impact of *Bt* crops impact on non target organisms at different trophic levels, an acquaintance with the majority of arthropod species prevalent in a given agro-ecosystem must be known.

Outbreak of secondary insect pests in *Bt* cotton : The outbreak of secondary pests in cotton is suspected because once the primary pest is brought under control, secondary pests have a chance to emerge due to the lower pesticide applications in *Bt* cotton cultivars. According to the studies conducted recently on emergence of secondary pest, more pesticide sprayings are needed over time to control emerging secondary pests, such as aphids, spider mites, and lygus bugs. Worldwide there are studies where *Bt* cultivars has proved their efficacy against target pests but their impact on the environment is still uncertain (Qaim and Zilberman, 2003).

It is premature to comment that adoption and cultivation of *Bt cotton* crop will not affect non target pests/secondary insect species due to the limited number of non target species studied (Lovei *et al.,*2009) available. However, previous studies on the side-effects of *Bt* toxins on non-target organisms yielded conflicting results; some researchers documented that the *Bt*-cotton has little or no effect on the non- target insects (Yunhe *et al.,*2011;Sujii *et al,* 2008; Liu *et al.,* 2006; Naranjo, 2005, Head *et al.,*2005) while others demonstrated that the population densities of non target insects such as cotton aphid, whitefly, and green leaf bug increased in cotton fields (Nguyen, *et al.,*2008; Fitt *et al.,* 1994) and no change in population densities of sucking insects or of the foliage feeder *Myllocerus undecimpustulatus* on Bollgard, Bollgard II and conventional cultivars (Dhillon and Sharma, 2013, Mann *et al.,* 2010). Considerable

information has been generated on the relative efficacy of *Bt* and conventional cotton in many countries (Tabashnik, *et al.,* 2008; Qaim and Zilberman, 2003) but reports regarding the performance of *Bt*-cotton in the tropics are few due to the recent adoption of *Bt* technology and also the tropics are often more complex and consist of several crops that serve as alternate and collateral hosts for bollworms and other sucking pests. This has lead to differences in performance of the different *Bt*-cultivars (Gopalaswamy *et al.,* 2009; Sharma and Pampapathy, 2006) in these countries.

The pest scenario in Indian cotton ecosystem is changing and adoption of *Bt* cotton has not only changed the cultivation profile, but also the pest scenario. While there is a decline in the pest status of bollworms; the sap feeders, viz. aphids, jassids, mirids and mealy bugs are emerging as serious pests (Akoijam *et al.,* 2014; Dhaliwal *et al.,* 2010; Vennila., 2008). Recently, mirid bugs, Ragmus spp. and *Creontiades biseratense* (Distant) appeared in epidemic form in Dharwar (Karnataka) and Coimbatore (Tamil Nadu). Mirid bug had appeared in severe form throughout the Karnataka, with most aggressive status in Haveri district (Manohar *et al.,* 2012; Rohini *et al.,* 2009; Udikeri *et al.,* 2009). Also, some of the minor pests like thrips, *Thrips tabaci* Linderman; shoot weevil, *Alcidodes affaber* Aurivillius and stem weevil, *Pempherulus affinis* (Faust) are becoming serious on *Bt* cotton (Sarode *et al.,* 2009). Various species of mealy bugs have started appearing in serious proportions on field crops, vegetables, fruits and ornamentals (Tanwar *et al.,* 2007). In fact, mealy bugs have become indicator insects for the current ecosystem alterations due to slow changes in climate during the period from 2002 to 2005. Among these, *Phenacoccus solenopsis* Tinsley on cotton and *Paracoccus marginatus* Williams and Granara de Willink on papaya have become quite serious (Rajendran, 2009; Tanwar *et al.,* 2007). During 2006, *P.solenopsis* appeared for the first time on cotton crop in Punjab and caused severe losses in some pockets of Ferozepur, Muktsar and Bathinda districts (Dhawan and Saini, 2009). Since then this pest has spread to several states like Haryana, Rajasthan, Maharashtra and Gujarat and southern states (Atwal and Dhaliwal, 2009; Nagrare *et al.,* 2009; Monga *et al.,* 2009). As Bollgard (Cry1Ac) cotton does not provide protection against tobacco caterpillar, *Spodoptera litura* (Fabricius), it continues to inflict heavy losses in several cotton growing regions of India (Dhaliwal *et al.,* 2010). New reports of damage by tea mosquito, *Helopeltis bradyi* (Waterhouse) and gall midge, *Dasineura gossypii* damage in Karnataka on MRC 6918 and RCH 708 *Bt* cottons are of concern (Kumar and Gopalaswami, 2014). A severe outbreak of whitefly has been observed in the north cotton growing zone of India during 2015.

Factors associated with secondary insect pests' outbreak in *Bt* cotton crop : The employment of *Bt* crops might have non nutritive negative effects on agricultural ecosystem interactions and on farm profits (Sharma and Ortiz, 2000; Wolfenbarger and Phifer, 2000).Secondary pests, which before were of minor importance, might now find favorable conditions and themselves become major pests (Lu *et al.,* 2010). Three main factors responsible for an outbreak of secondary pest species with the use of *Bt* cotton are (i) reduction in application of broad-spectrum insecticide for target pests; (ii) impact on natural enemy populations; or (iii) reduction in interspecific competition due to the absence of primary/target pest. Additionally under Indian conditions the cultivation of large number of *Bt* cotton

genotypes (hybrids) may also be having an important role in secondary pest outbreak.

Genotypic reaction : By May 2012, there were 1128 *Bt* cotton hybrids available in the Indian market (IGMORIS, 2012). *Bt* cotton currently occupies over 93 per cent of the area under cotton cultivation. Genetic makeup of the plant is very much important to confer tolerance to biotic and abiotic stress under natural conditions. Sucking pests have emerged as major pests causing significant economic losses for which one of the possible reasons is that the donor parent Coker 312 itself is highly susceptible to sucking pests (Kumar and Gopala Swamy,2014).Furthermore these hybrids escaped the rigorous screening procedure against insect-pests prior to approval. 54 *Bt* cotton hybrids were evaluated for genetic tolerance to sucking pests and leaf reddening under rain fed farming, where differential reactions in hybrids to sucking pest recorded, providing an option for cotton stake holders to choose tolerant hybrids so that indiscriminate insecticide sprays can be reduced (Nagrare *et al.,*2014). The hybrids released for cultivation has different plant type, morph-physiological traits, agronomic requirement and the microclimate of the crop is disturbed if these conditions are not followed.

Reduction in broad spectrum insecticide applications in *Bt* cotton and secondary pests outbreak : Due to its efficacy against target pests , introduction of *Bt* technology brought significant decreases in insecticide application among adopters, at least during the early years and considerably alleviated the negative impacts associated with the use of such insecticides (Krishna and Qaim, 2012; Kouser and Qaim, 2011; Birader and venilla, 2008). Despite warnings from several authors (Sharma and Ortiz, 2000; Wu and Guo, 2005) that some secondary insect pests could appear in such numbers that they become key pests in *Bt* crop fields, specific measures to overcome their population increases were not taken. Consequently, there have been outbreaks of secondary pests which were previously controlled by the insecticide applications originally targeting the primary pest (Prasad *et al.,* 2014; Kranthi, 2012; Pemsl *et al.,* 2011; Lu *et al.,* 2010). This situation has been particularly evident in *Bt* cotton production in China. Less than 3 years after its introduction in 1998, several pest groups including whiteflies, plant hoppers, aphids, mirids and mealy bugs increased in number (Yang *et al.,*2005a; Men *et al.,* 2004). After the introduction of *Bt* cotton, cotton cultivators in India have been facing new problems with insect pest management in many parts of the country, mostly presumed to be a consequence of low insecticide usage. New sucking pests have emerged as major pests causing significant economic losses. Insecticide usage for bollworm control decreased after 2004 from 6454 to 222 metric tonnes and usage for sucking pest control increased after 2006 from 2735 to 6372 metric tonnes in 2011 (Kranthi, 2012). It is known that the usage of synthetic pyrethroid for bollworm control had significant negative impact on the incidental populations of *Spodoptera* sp. and several other miscellaneous bugs including the mirid bugs, *Creontiodes biseratence* (Distant) and Ragmus sp. The reduction of pyrethroid and several conventional insecticides on *Bt* cotton is presumed to have led to an enhanced infestation of several non target species such as mirid bugs, mealy bugs, thrips and tobacco cutworm (Kumar and Gopala Swami, 2014)

Association between reduction in natural enemies and secondary pests' outbreak in *Bt* cotton crop : Natural enemies are critical to ecosystem functioning by inhibiting the excessive multiplication of potential pests in agricultural systems through 'biological control' (Bianchi *et al.,* 2006; Wilby and Thomas, 2002). Natural enemies alone may be sufficient in some cases to keep secondary pest populations under economic injury thresholds (Wolfenbarger *et al.,* 2008; Hardin *et al.,* 1995).The employment of *Bt* crops and the consequent reduction in insecticide usage increase the significance of the function of natural enemies to control secondary pests (Naranjo, 2005).Hence, a major concern related to the growing of *Bt* crops is their potential impact on the abundance of natural enemies (Marvier *et al.,* 2007; Poppy and Sutherland, 2004). Predators were less abundant in *Bt* cotton fields compared to unsprayed non *Bt* control fields. Fewer specialist parasitoids of the target pest occurred in *Bt cotton* crop fields compared to unsprayed non *Bt* controls, but no significant reduction was detected for other parasitoids. Numbers of predators and herbivores were higher in *Bt* crops compared to sprayed non *Bt* controls (Wolfenbarger *et al.,* 2008).

The selectivity of Cry toxins is not entirely known, with the potential for unintended effects on beneficial species which may influence other non susceptible pests (Lovei *et al.,* 2009). However, interactions between prey and natural enemies are extremely complex. Many laboratory and field research studies evaluated the impact of *Bt* toxins on the natural enemies of potential secondary pests. Several laboratory studies reported no significant effects on natural enemies (Andow *et al.,* 2009) while others have indicated negative effects (Dhillon and Sharma., 2009; Gonzalez-Zamora *et al.,* 2007). Results from studies performed at a field level show similar variation; some found no significant impacts while other reported negative effects. Natural enemies are often present in higher numbers in insecticide-free conventional fields than on *Bt* fields (Naranjo, 2009; Marvier *et al.,* 2007). It is also widely accepted that the use of insecticides has larger direct negative effects on natural enemies than does the use of *Bt* crops (Romeis *et al.,* 2009; Wolfenbarger *et al.,* 2008; Cattaneo *et al.,* 2006). Overall, this suggests that in field settings, where *Bt* cotton crops do have an impact on natural enemies, but these are not as strong as the direct effect of insecticide. The reports indicating both negative and positive effects of cry toxins on natural enemies need further detailed and long term laboratory and field studies.

The impact of *Bt* toxins on natural enemies can occur through direct and/or indirect effects (Romeis *et al.,* 2006).

Direct impacts might occur due to the ingestion of the insecticidal protein. The mechanism of action of several available *Bt* toxins is still unknown or in conclusive (Lovei *et al.,* 2009; Lovei and Arpaia, 2005) leading to the assumption that *Bt* toxins may cause similar negative effects on predators as they do on the target herbivores (Andow *et al.,* 2006).

Manifestations of Indirect effects might be through reductions in prey/host populations or in the nutritional quality of the prey. Impacts of the toxin on herbivores may manifest at a sub lethal level which can affect life parameters such as life span and fecundity (Romeis *et al.,* 2004). There is evidence that the low nutritional quality of prey items after they have ingested *Bt* proteins has a significant impact on the performance, development and even survival of natural enemies. Moreover, high mortality rates

in the target species may cause a reduction in specialist natural enemies, which themselves can be important prey for generalist predators. Additionally, prey species in general might migrate to non-*Bt* fields in search of preferable food resources. Thus, if prey availability for secondary pest predators in *Bt* fields is scarce, predators might be encouraged to 'migrate' to adjacent conventional crops, negatively affecting their abundance within *Bt* fields (Sisterson *et al.,* 2007). Hence, it may be possible that these negative impacts will permit the development of secondary pests in the crop itself or even in neighboring crops (Gross and Rosenheim, 2011; Gutierrez *et al.,* 2006). A clear and strong understanding about the direct and indirect effects of *Bt* cultivars on natural enemies is important as these insects play a major role in biological control of primary as well as secondary pests.

Secondary insect pests' outbreak due to species replacement : Competition may play an important role in the dynamics of herbivorous insects (Kaplan and Denno, 2007) and cotton crop has so far been reported to be attacked by 1326 insect species (Hargreaves 1948) where the phenomenon of species replacement become highly relevant. However, the importance of replacement between primary and secondary pests has generally been ignored in conventional agriculture (Denno *et al.,* 1995; Hardin *et al.,* 1995) and especially in *Bt* cropping. *Bt* cotton crop, similar to insecticides, is an artificially imposed disturbance on the ecosystem; hence, it is not surprising that niche rearrangement occurs. It is possible that when a primary pest is successfully controlled by a *Bt* toxin, a non-susceptible species starts to utilize the newly available ecological resource (Gross and Rosenheim, 2011; Hardin *et al.,* 1995). This situation occurs in cases where, prior to the pest management treatment, the primary pest is a dominant competitor species and the secondary pest is a weak competitor (Shivankar *et al.,* 2007).

In *Bt* cotton in the USA, stink bug pests, specifically *Nezara viridula* L. and *Euschistus servus* S., have recently become a severe problem in the absence of the target pests *Heliothis zea* and *H. virescens* (Zeilinger *et al.,* 2011). As *Bt* cropping expands worldwide; it is of critical importance to determine the key species–susceptible and non susceptible pests–which might compete for resources within the same *Bt* crop. An increase in abundance of tobacco caterpillar, *Spodoptera litura* in bollgard cotton in North India during 2005-06 was due to suppression of primary bollworm, *Helicoverpa armigera* .

Secondary pests' outbreak and its impact on *Bt* cotton crop : In the early years of *Bt* cropping, there were reports of increased profitability in overall production due to 40%–60% reductions in insecticide applications alongside increased crop yields, as compared to non- adopters (Rui *et al.,* 2005; Bennett *et al.,* 2004; Qaim and Zilberman, 2003; Huang *et al.,* 2002; Pray *et al.,* 2002; Fitt, 2000). Nonetheless, there were early concerns about the potential for secondary pest outbreaks due to the decrease in insecticide applications (Morse *et al.,* 2005; Qaim, 2003; Wu *et al.,* 2002).

Cotton From the worldwide 24.3 million hectares cropped with *Bt* cotton, India, China and USA account for 11.0, 4.2 and 4.1 million hectares, respectively (James, 2013), with very high adoption rate (James, 2013). Until the end of the 20th century, insecticides were

intensively applied to control the cotton bollworm (Wu and Guo, 2005). However, in the early 1990s, the effective control of this pest became problematic, as cotton bollworms became resistant to most insecticides due to their repetitive and overuse (Deguine *et al.,* 2008; Wu and Guo, 2005; Kranthi *et al.,* 2002,).

In China after the introduction of *Bt* technology in 1999, insecticide applications in *Bt* cotton fields dropped from about 61 kg/ha (20 applications) per year, to approximately 12 kg/ha (6.6 applications) per year (Huang et al ,2002). By 2002, this figure started to increase, reaching on average 15.6 kg/ha (10.7 applications) per year of insecticides, of which 4.7 kg were used against cotton bollworm, and the remaining against lygus bug and other pests (Pemsl et al, 2011). By 2005, farmers applied roughly the same amount against the cotton bollworm, but the amount sprayed against secondary pests had increased by 20 per cent, to a total of 18.6 kg/ha (14.2 applications) per year (Pemsl et al 2011). The drop in insecticide use and the ineffectiveness of *Bt* cotton against these secondary pests has led to a reversal of the ecological role of cotton (Li *et al.,* 2011; Lu *et al.,* 2010).Conventional cotton had been a population sink for the miridbug secondary pest, while now a day's *Bt* cotton fields are a source of these pests (Lu *et al.,* 2010,) and in India the sap feeders, *viz.,* aphids, jassids, mirids and mealy bugs are emerging as serious pests (Akoijam *et al.,* 2014; Dhaliwal *et al.,* 2010; Vennila, 2008). This has led to a situation where there are no major differences in the total quantity and expenditure in insecticide application between *Bt* and conventional cotton farmers (Zhao *et al.,* 2011). In India, usage of insecticides for sucking pest control increased after 2006 from 2735 to 6372 metric tons in 2011 (Kranthi, 2012).

But according to Huang *et al* 2014, cotton production is still effective and farmers are applying fewer sprayings in early season, with fewer cases of human poisoning. Moreover, a higher survival of generalist arthropod predators has been recorded (ladybirds, lacewings and spiders), providing additional biocontrol to neighboring crops, such as maize and soybean (Huang *et al.,* 2014; Lu *et al.,* 2012).

Indian cotton farming is comparable with China, with numerous small-scale farmers (Qaim *et al.,* 2009; Huang *et al.,* 2002;).Recent evidence showed that secondary pests are now posing a major problem (Nagrare *et al.,* 2009), with farmers battling against non target insects (Ramaswami *et al.,* 2012; Stone, 2011) found no significant difference between adopters and non-adopters in terms of insecticide use. Elsewhere in the world, similar issues to the Chinese and Indian cases have been reported in cotton. Adopting farmers are either still using significant numbers of insecticide applications to control secondary pests, or the damage caused by these pests has increased. Some examples include South Africa (Schnurr, 2012; Hofs *et al.,* 2006), Pakistan (Jaleel *et al.,* 2014), Australia (Wilson *et al.,* 2013), Brazil (Sujii *et al.,* 2013) and Mexico (Traxler and Godoy-Avila, 2004). The development of resistance in target pests and appearance of secondary pests has led to increase in number of applications to control these pests in cotton and quantity of insecticides used against these pests is also increasing pointing towards a more robust insecticide resistance management program for overall sustainability of cotton ecosystem.

CONCLUSION

The secondary pests are a matter of serious concern and need to be addressed with greater emphasis. The cotton crop worldwide has

been reported to be attacked by large number of insect-pests species and there are chances of competitive displacement or acquisition of vacant niche by non competent species either in the absence of primary insect–pest species or due to reduction in insecticidal application or changes in natural enemies' scenario.

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Bt cotton and pest management : Expliotation of insecticides for pest management

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Cotton crop is cultivated in 120 countries across the five continents over 23.9 million hectares. India, China, USA and Pakistan are the major cotton growing countries. The numerous insect species from sowing to maturity in cotton growing area of the world and damage by these pests is major constraints in productivity. Management of these pest is mainly based on use of insecticides. The over exploitation of insecticides has resulted in volatile cotton ecosystem and heavy losses occur in one or other cotton growing area every year mainly due to failure of insecticides dominated integrated pest management. The failure of insecticides due to development of insecticides resistance to key pests (jassid, whitefly and bollworms) shattered the confidence of cotton growers in cultivation of cotton crop. Development of resistance to insecticides to bollworms particularly *Helicoverpa* resulted in adoption of *Bt* cotton in 1996. At present more than 70 percent area is under transgenic cotton and providing effective control of bollworm complex. They pest complex has changes after introduction of *Bt* cotton in cotton growing area of the world. There may be several ecological impacts induced by transgenic cotton due to change in biodiversity, susceptibility of *Bt* cotton hybrids to various insect pests, change in tritrophic interaction, apart from their direct impact on target pest (bollworms and other lepidopteran pests like leaf folder, semilooper) . The impact of *Bt* cotton on plant defence system need through investigation on short and long tern basis. The interactions between target insect and transgenic cotton, and the way of toxic expressed by transgenic cotton varied with plant spatial parts and different growing stages on are regarded as the main cause for insect to develop resistance. The insect community structure including insect pests and beneficial organisms is less stable in *Bt* cotton than that in non *Bt* cotton . It is much easier for minor insect pests developing to be major pest. In Indian sub continent ,before the introduction of *Bt* cotton, 8 were the key pests which cause major damage to the crop . Among these, cotton jassid [*Amrasca biguttula* (Ishida)], whitefly [*Bemisia tabaci* (Gennadius)], pink bollworm [*Pectinophora gossypiella* (Saunders)], spotted bollworms [*Earias vittella* (Fabricius) and *E. insulana* (Boisdual)], American bollworm [*Helicoverpa armigera* (Hubner)] are pest of national importance. The thrips [*Thrips tabaci* Lindman] cause moderate to heavy damage in Central zone (Dhawan *et al.* 2011, 2012). With introduction of *Bt* cotton, bollworms are not serious pests as *Bt* cotton provides effective control of this group of pests. The jassid damage has increased and new sucking pests like mealy bugs, and mirid bugs damaged the cotton crop in recent past (Dhawan *et al.,* 2009). The tobacco caterpillar is another new pest that is has gained importance. But with introduction of Bollgard II, the status has declined to minor pest. Failure of *Bt* cotton due to whitefly in north irrigated *hirsutum* cotton

(India) during 2015 is warning for rest of the cotton growing area for emergence of outbreaks of minor pests.

Emergence of secondary pests : The adoption of *Bt* cotton resulted in increase of non lepidopteran pests mainly sucking pests over time as relatively large number of pest species that are not susceptible to the *Bt* toxins expressed in transgenic cottons and this has affected cotton production worldwide. The sucking pests, including the cotton aphid, thrips, whitefly, mealy bugs, leafhopper, stink bug, red cotton bug, dusky cotton bug and spider mites, are the major non-target pests in *Bt* cotton fields, which are not susceptible to the *Bt* proteins currently used. Due to the reduced use of insecticides for bollworms management and the change of pest management regimes in *Bt* cotton fields as result of change in biodiversity of cotton ecosystem, these secondary pest populations have increased and evolved into key pests in the USA, India, Pakistan, China, Australia, and other countries (Gouse *et al.,* 2004; Williams 2006; Wilson *et al.*, 2006; Ho and Xue 2008; Lu *et al.*, 2008; Li *et al.*, 2010; Zhao *et al.,* 2011, Dhawan and Bal 2007, Dhawan and Saini 2007, Dhawan *et al.,* 2007, 2011,2012). The insecticides for management of bollworms control provided effective control of sucking pests during reproductive phase of cotton crop . In short, to cultivation of *Bt* cotton, secondary pests have a chance to emerge and without additional pest control, could potentially evolve into primary pests . Several studies in different cotton growing of world have predicted the emergence of secondary pests (Wang *et al.,* 2008; Xu *et al.*, 2008). Many of sucking pests (mirids, aphids, stink bugs, mealy bugs, thrips, cotton stainers etc.) are most likely to show such effects in different cotton growing areas of world. In Australia, the green mirid (*Creontiades dilutus*), green vegetable bug (*Nezara viridula*), leaf hopper (*Austroasca viridigrisea* and *Amrasca terraereginae*), and thrip (*Thrips tabaci, Frankliniella schultzei* and Frankliniella occidentalis) have become more prominent (Lei *et al.*, 2003; Wilson *et al.,* 2006). In China which has adopted *Bt* cotton in large scale, the increase in mirid infestation is largely related decreased pesticide use due to *Bt* cotton (Lu *et al.,* 2010) and fluctuations in population to local temperature and rainfall (Huang *et al.,* 2010). The transgenic crops did not contribute to the nontarget pest outbreaks (*Adelphocoris suturalis* Jakovlev.) when being compared with their parental lines in China. The possible reasons for intensified pest status of *A. suturalis*, were decrease of pesticide application, deficient natural enemies, and area-wide shift of cotton varieties (Guoping *et al.,* 2010). In India, mirid bug ,mealy bugs and whitefly emerged as new pests on *Bt* cotton (Dhawan *et al.,* 2007, 2011; Dhawan and Kumar, 2013).

The sucking pests have become a more significant part of the pest complex in *Bt* crops in some countries and additional spraying are required where the advantage of the *Bt* crop has been significantly eroded. In the south-eastern USA, stink bugs have assumed significant pest status in *Bt* cotton crops (Greene *et al.* 2001). The removal of broad spectrum sprays directed against *Helicoverpa* spp has resulted in higher densities of plant bugs (*Creontiades* spp) and stink bugs (*N. viridula*), and to a lesser extent leafhoppers (*Amrasca terraereginae* (Paoli). In India, mealy bugs, thrips, mired bugs, leaf-eating caterpillar and recently whitefly has emerged as potential threat to cultivation of *Bt* cotton (Karihaloo and Kumar 2009; Nagrare *et al.*, 2009). Field surveys conducted over 10 years in six major cotton-growing provinces (*i.e.*, Henan,

Hebei, Jiangsu, Anhui, Shangdong, and Shanxi) in northern China showed that mirid bugs (Heteroptera: Miridae) have progressively increased and acquired pest status in *Bt* cotton fields (Lu *et al.*, 2010). In addition, that spider mites have been observed to occur at higher levels in *Bt* cotton during the drought season (Wu and Guo 2005). In Australia, reduction in sprays applied for *Helicoverpa* was accompanied by no change in sprays for mirids, aphids, mites and thrips (Fitt 2004). *Bt* cotton in China help to prevent resurgence of aphid populations (Wu and Guo 2003). However, the emergent pests have forced cotton farmers to continue using chemical pesticides; however, the increase in insecticide use for the control of these secondary insects was initially lower than the reduction in total insecticide use due to *Bt* cotton adoption (Wang *et al.*, 2009) but now has increased substantially. With introduction of *Bt* cotton in Punjab, incidence of jassid and whitefly is on increase. In north cotton belt of India (Punjab, Haryana and Rajasthan) whitefly emerged as serious pest in 2015 which resulted in 30-60 loss to cotton crop in spite of increase in number of sprays to 7-10. This has shattered the economic of *Bt* cotton and warning for cotton growing countries to have strategic planning for management of emerging pests problems. *Bt* cultivars are one of the main reason for emergence of new pests.

The improved pest management strategies-integrating *Bt* with different methods of pest management are needed to control secondary pests. However, at present scenario are dependent on use of pesticides which increased the cost of production and excessive use may result in emergence/resurgence of secondary pests. In present scenario additional sprays required for management of regular sucking pests.

Pesticide use : The massive publicity for the use of insecticides for insect pest management programs in cotton resulted in exploitation of insecticides as only solution to insect pest problems in cotton. This has resulted in failure of good insecticides like synthetic pyrethroids and chloronicotnyl (CNC) for management of cotton pests. The replacement with new insecticides resulted in higher cost of pest management in cotton. The non chemical techniques, such as cultural control, , resistant cultivars, and biological control etc are widely are advocated as first line of defence against insect pests and insecticide should be used primarily as a last line of defence. However, cotton pests cannot be controlled adequately without some insecticide use. Recent failure of cotton crop due *Helicoverpa* is result of development of resistance to insecticide particularly synthetic pyrethroids most common used insect world wide. The pyrethroids were developed which were generally applied at lower rates and did not bioaccumulate. But being less persistent, it often was necessary to apply them more frequently. Similarly, the jassid (*Amrasca biguttula*)developed resistance to chloronicotnyl particularly imidacloprid due to use as seed treatment. This is due the seed treatment of *Bt* cotton which is marketed . This has resulted in development of resistance to jassid and field dose of CNC is 2-3 times more which was initially recommended. This will further increase cost of cultivation. Moreover, in north India, the seed treatment has no additional benefit as spray against this pest starts in July irrespective of seed treatment or not. It was obvious that chemical-based pest management programs could not be maintained and that, in future, acceptable levels of pest control would be achieved only through better combination of chemical and non-chemical management

techniques. Recently, many advances have been made in developing these integrated pest management techniques particularly for insect control. The *Bt* cotton provided effective control of bollworm complex on cotton and the use of insecticides for this group of lepidopteran is negligible as no serious reports of resistance has been reported . There are certain report on the development of resistance to Cry 1 Ac. The stacked *Bt* containing Cry 1 Ac and Cry 2 Ab also provided effective control of *Spodoptera* which emerged as new pest after introduction of *Bt* cotton in India. The reduction in insecticides and number of sprays was one of the aim of *Bt* cotton . On average, the pesticide sprayings had decreased five to six times in growing areas these pests . This has also reduced number of sprays on non *Bt* cultivated in vicinity of *Bt* cotton. The incidence of insect pests common on alternate crops has also reduced in cotton belt. *Bt* cotton is only effective against the bollworms, while secondary pests have increased is well documented. *Bt* crops not only provide an effective alternative tool for controlling target insects (Wu *et al.*, 2008), but also provide many social, environmental, and economic benefits, such as reducing the use of chemical insecticides, benefiting the environment and human health, and increasing farm income (Choudhary and Gaur 2010; Huang *et al.*, 2010;). Nevertheless, as with any technology, there have been questions about the potential risks transgenic plants might have on the environment. One of the major ecological concerns regarding the environmental risks of insect-resistant GE plants is their potential effects on non-target organisms (NTOs)

Cotton industry and managing pesticides use : Without effective management programs, losses caused by insect pests can result in uneconomical cotton production would. The management of insecticide resistant and losing effective chemicals one after another and environmental concerns, has been a constant challenge for scientific institution and cotton growers. Even now current cotton pest management programs still rely heavily on chemical use and. pesticide management research is not priority. This led to a number of problems, making cotton cropping ecosystem as more fragile result in failure due to one or other insect pests. Failure of cotton due to whitefly in north irrigated cotton during 2015 is result of insecticides based pest management strategy.

In managing pesticide use, there are three logical steps: · minimize insecticide input, selection of pesticides with minimal environmental impact and adoption of ecological approach for suitable insect pest management. The .conventional insecticide use need to be curtailed. Reduction of pesticide use has been the priority of scientists for many years, because it is important in terms of managing insecticide resistant pests, resurgence of new insect, loss of biodiversity. Through development of insect monitoring programs, economic thresholds and other techniques, substantial progress has been made but there are many constraints particularly in developing and under developed cotton growing countries. The appearance new pest problem and minor insects into major inset pest further increased the use of insecticides. Pesticide is still essential to cotton production post *Bt* adoption. However, a few study indicate that farmers are using nearly 3 times more pesticide than the optimal profit maximizing level

Newer insecticides and pest management : The new chemistry has advantages as it has lower mammalian toxicity(short re entry and pre harvest intervals, allowing

the insecticides to be easily incorporated into pest control programs), less resurgence problem, short restricted entry intervals (REIs) , environmental protection, and pest management selectivity (greater selectivity to target specific species, so they are less likely to harm natural enemies).

1. Neonicotinoids : Neonicotinoids are the most effective insecticides for the control of sucking insect pests such as aphids, whiteflies, leaf hoppers, thrips, and some micro lepidoptera and a number of coleopteran pests. Their broad spectrum of efficacy, together with systemic and translaminar action, pronounced residual activity and a unique mode of action, make the neonicotinoids the most rapidly expanding insecticidal class since the launch of the first compound, imidacloprid, by Bayer Crop Science in 1991. Six additional neonicotinoid insecticides were launched: nitenpyram (Sumitomo Chemical Takeda Agro Company, 1995), acetamiprid (Nippon Soda, 1995), thiamethoxam (Novartis, 1998), thiacloprid (Bayer Crop Science, 2000), clothianidin (Sumitomo Chemical Takeda Agro Company, Bayer Crop Science, 2000) and dinotefuran (Mitsui Chemicals, 2002) . They act on the central nervous system of insects. Their action causes excitation of the nerves and eventual paralysis, which leads to death. Because they bind at a specific site (the postsynaptic nicotinic acetylcholine receptor), they are not cross-resistant to the carbamate, organophosphate, or synthetic pyrethroid insecticides, which was an impetus for their development.

(a) Imidacloprid: Imidacloprid is a systemic neonicotinoid insecticide and upon the nervous system, causing blockage of postsynaptic acetylcholine receptors. Because of the systemic mode of action and low toxicity to humans, imidacloprid has become a popular insecticide worldwide for use in ornamentals, field crops and vegetables and is registered in approximately 120 countries and is used on over 140 different agricultural crops. In the field, imidacloprid used as a seed treatment, is linked to alteration in foraging, recruitment, and mortality of honey bee, *Apis mellifera* (L.).

(b) Acetamiprid: Acetamiprid is a systemic neonicotinoid insecticide developed by M/s Nippon Soda Co. Ltd., for use against sucking insects, such as jassid, aphids and whiteflies on cotton. It acts on the central nervous system causing irreversible blocking of the postsynaptic nicotinergic acetylcholine receptors Acetamiprid degrades rapidly and poses low risks to the environment

(c) Thiacloprid : Thiacloprid is a neonicotinoid insecticide with systemic properties It has activity not only against sucking insects such as aphids, whiteflies, and jassids but is also active against weevils, leafminers, On the basis of its high insecticidal activity with a favourable ecological profile and safety to bees will be useful in cotton ecosystem. Like other chloronicotinyl insecticides, thiacloprid acts selectively on the insect nervous system as an agonist of the nicotinic acetylcholine receptor (nAChR).

(d) Thiamethoxam : Thiamethoxam is a novel neonicotinoid belonging to sub class of thianicotinyl compounds and it represents the first example of second generation neonicotinoids with a unique structure and outstanding insecticidal activity introduced by Novartis . This is a systemic insecticide for soil and foliar applications and control a variety of

pests such as aphid, whiteflies, thrips, betles, leaf hopper. Thiamethoxam's chemical structure is slightly different than the other neonicotinoid insecticides, making it the most water soluble of this family. Because of its greater water solubility, it moves readily in plant tissue The compound shows contact as well as exceptional systemic activity.

(e) Clothianidin: Clothianidin is a novel, highly effective systemic and contact insecticide exhibiting low mammalian toxicity. Clothianidin is active on hemipteran pest species in particular, such as aphids, leafhoppers and also on many coleopteran and some lepidopteran pest species with a low application. Like other neonicotinoid insecticides, clothianidin also acts as an agonist at the nicotinic acetylcholine receptors (nAChR) located in the central nervous system

(f) Dinotefuran: Dinotefuran is a new neonicotinoid developed by Mitsui Chemicals and has a characteristic tetrahydro-3-furylmethyl group instead of the aromatic heterocyclic ring that was previously considered indispensable for insecticidal activity of neonicotinoids. It has excellent insecticidal properties and offers excellent control of a wide variety of pests in many kinds of crops . particularly hemiptera, lepidoptera, and thysanoptera. Dinotefuran acts through contact and ingestion and results in the cessation of feeding within several hours of contact and death shortly after. Dinotefuran was granted Organophosphorus Alternative and Reduced Risk Status by the EPA. Dinotefuran is water-soluble and has excellent systemic and translaminar action in many plants.

2. Phenyl pyrazole

(a) Fipronil : Fipronil is a phenyl pyrazole insecticide first synthesized by M/S Rhône Poulenc Ag Company (now Bayer Crop Science) .. It has contact, systemic and ingestion activity. Fipronil is applied at low doses, is effective against cotton (thrips). Fipronil act by interfering with the passage of chloride ions through the ã-aminobutyric acid regulated chloride channel, thereby disrupting central nervous system activity and causing death. This causes the over-excitation of the muscles and nerves of infected insects, causing them to death. It belongs to toxic category of pesticides.

3. Oxadiazines

(a). Indoxacarb: Indoxacarb is the first commercialized insecticide of the oxadizine and is a mixture of R and S isomer in the ratio of 25:75. The active isomer is S isomer. This insecticide has a novel mode of action and acts by inhibiting sodium ion entry into nerve cells, resulting in paralysis and subsequent death of pests. It acts primarily as a stomach poison as well as by contact action. Indoxacarb is effective against lepidopteran pests of cotton and is considered a reduced risk pesticide with low mammalian toxicity and a benign profile for avian and aquatic toxicity as compared to that of conventional insecticides.

4. Pyrrole

(a) Chlorfenapyr : Chlorfenapyr is the first commercial pesticide to be derived from a class of microbially produced compounds known as halogenated pyrroles from a naturally-produced chlorinated pyrrole, Chlorfenapyr is a 'proinsecticide', *i.e.,* it requires activation through metabolism. The parent compound is converted to a metabolite, which functions as an uncoupler of oxidative phosphorylation at mitochondria. It has broad spectrum activity against many species of bollworms and

thysanoptera . It is mainly a stomach toxicant, but has some contact activity. Chlorfenapyr works by disrupting the production of adenosine triphosphate. resulting in disruption of production of ATP, cellular death, and ultimately mortality.

5. Pyridine azomethine

(a) Pymetrozine : Pymetrozine a novel pyridine azomethine insecticide, is highly specific against sucking insect pests. Its unique mode of action is not yet entirely understood, but it affects the nerves controlling the salivary pump and causes immediate and irreversible cessation of feeding due to an obstruction of stylet penetration, followed by starvation and insect death. The compound is a powerful toxicant against aphids [*Aphis gossypii* Glover } and whitefly [*Bemesia. tabaci* }. Pymetrozine has systemic and translaminar activities and can be used as drench or foliar application . The compound appears to have great promise in IPM programmes due to its high degree of selectivity, low mammalian toxicity, and safety to birds, fish and non target arthropods.

6. Diamides

(a) Flubendiamide : Flubendiamide belongs to phthalic acid diamide developed by Bayer Crop Science, Germany in collaboration with Nihon Nohyaku Co. Ltd., Tokyo, Japan and introduced in many countries during 2005-2007. It has a novel biochemical action as it affects calcium ion balance irrespective of sodium or potassium ion balance, which causes contraction of insect skeletal muscle. Flubendiamide is mainly effective for controlling lepidopterous pests including *Helicoverpa spp, Spodoptera spp*

(b) Chlorantraniliprole : Chlorantraniliprole, belongs to a new chemical class, the anthranilic diamides, and has a novel mode of action as an activator of insect ryanodine receptors, causing rapid muscle disfunction and paralysis. This insecticide is effective against bollworms complex of cotton. Chlorantraniliprole is classified as a reduced-risk pesticide. The key biological attributes of chlorantraniliprole are its selectivity profile towards non-target organisms and its long-lasting activity against target pest species particularly in term of the speed to cause

Table 1. Classification of newer insecticides

Sr No.	Class of the Insecticides	Name of the insecticides
1.	Neonicotinoids	Imidacloprid, Acetamiprid, Thiacloprid, Thiamethoxam, Clothianidin, Dinotefuran,
2.	Phenyl pyrazoles	Fipronil
3.	Oxadiazines	Indoxacarb
4.	Pyrrole	Chlorfenapyr
5.	Pyridine Azomethines	Pymetrozine
6.	Diamide	Flubendiamide, Chlorantriniliprole
7.	Pyridine carboxamid	Flonicamid
8.	Tetronic Acid Derivatives	Spiromesifen
10.	Tetramic Acid Derivative	Spirotetramat
11.	Bacterial Fermentation Products	Spinosyns,
12.	Dichloropropenyl Ethers	Pyridalyl
13.	Benzoylphenyl urea	Novaluron, Buprofezin

cessation of feeding .

7. Pyridine carboxamid

(a) Flonicamid: Flonicamid, a novel compound with a unique mode of action, has contact activity and is upwardly systemic. Flonicamid effectively manages populations of aphids, thrips, leafhoppers, and other sucking insects on cotton. It has no negative impact on beneficial insects or natural enemies. This compound acts as an antifeedant and a behavioral modifier resulting in insect mortality from starvation within 5 to 7 days after exposure. Flonicamid inhibits feeding by inhibition of stylet penetration to the plant tissue. The feeding disruption appears to be related to the neurological effect of flonicamid, resulting in death from starvation.

8. Tetronic acids derivative

(a) Spiromesifen : Spiromesifen is a novel insecticide/acaricide belonging to the new chemical class of spirocyclic phenyl-substituted tetronic acids, recently introduced by Bayer Crop Science, Spiromesifen is especially active against whiteflies (*Bemisia* spp). It has new mode of action that leads to the inhibition of the fat synthesis and acts as lipid biosynthesis inhibitor. Spiromesifen has a low toxicity to honeybees and bumble bees and extremely effective against pyriproxyfen and neonicotinoid resistant Q type whiteflies. Therefore it is an excellent resistance management tool in combination/rotation strategies. Spiromesifen is considered as harmless (category 1) to parasitoid *Eretmocerus mundus* Mercet.

9. Bacterial fermentation product

(a) Spinosad : Spinosad is active on various insect pests, especially bollworm complex and thrips and termites. It acts by depolarizing insect neurons involving acetylcholine and GABA receptors. Spinosad is classified as reduced risk product due to its unique mode of action, coupled with high degree of activity against targeted pests, low toxicity to mammals, fish, birds, wildlife and human beings, and of course, beneficial arthropods and may be used as an alternative to conventional insecticides in integrated pest management program. Due to its safety profile and biosynthesis during fermentation of *S. spinosa*, spinosad has been classified as bioinsecticide. Spinosad exhibits wide margins of safety to many beneficial insects.

(b) Emamectin benzoate: 'Emamectin benzoate is a novel non-systemic insecticide and is derived from a natural fermentation product, avermectin $B_{1.}$ It is highly toxic to a broad range of lepidoptera pest species (bollworm complex and *Spodoptera* spp) at very low concentration and penetrates leaf tissues by translaminar movement. It is neuroactive affects ion transfer through cell membranes (chloride channel activator). The avermectins act by disrupting nerve impulses by a unique mode of action It paralyses the lepidoptera larvae, which stop feeding within hours of ingestion, and die 2-4 days. Avermectin B1 (abamectin) is nother semi-synthetic avermectin derived from fermentation of avermectin B1.

10. Dichloropropenyl Ethers

(a) Pyridalyl: Pyridalyl is the only member of class propyl ether and has outstanding insecticidal efficacy toward *Helicoverpa armigera* and the tobacco cutworm, as well as against *Tysanopteran* insects.. This novel insecticide invented and developed by Sumitomo Chemical Co., Ltd. Pridalyl is environmentally friendly insecticide that is

suitable for use in IPM systems, as it possesses a high level of safety for humans, animals and fish and has minimal impact upon natural pest predators, such as parasitic wasps, pirate bugs (*Orius strigicollis*), lacewing, ladybugs, predatory mites and spiders, as well as on pollinating insects, such as the honey bee and the bumble bee .

11. Benzoylphenyl urea

(a) Novaluron : Novaluron is an insecticide of the benzoylphenyl urea class of insect-growth regulators (IGR). It is a relatively new chitin synthesis inhibitor, that inhibits the chitin formation. Novaluron is powerful suppressor of lepidopteran larvae such as *Spodoptera littoralis* and *Helicoverpa armigera* (Hübner) (by ingestion) and of cotton whitefly larvae *Bemisia tabaci* (Gennadius).

(b) Buprofezin : Buprofezin belongs to class thiadiazin and is good insect growth regulator effective against the nymph stages of whitefly, scales, mealybugs and leafhoppers by inhibiting chitin biosynthesis, suppressing oviposition of adults, and reducing viability of eggs. Buprofezin suppresses embryogenesis and progeny formation of *B. tabaci*. Non-systemic insecticide with contact and stomach action, and repellent properties and gives rapid knockdown and long residual activity.

12. Pyridine

(a) Pyriproxyfen: : It is a pyridine based pesticide which is found to be effective against a variety of arthropods and was introduced to the US in 1996 to protect cotton crops against whitefly. Pyriproxyfen is a juvenile hormone analogue, preventing larvae from developing into adulthood and thus rendering them unable to reproduce. It prevents them from maturing into reproductive adults and cause morphological abnormalities and disrupts the normal development of immature stages of insects, leading to sterility or death.

Exploitation of insecticides for insect pest management : The over use of insecticides alone and in combination for cotton pest management before introduction of *Bt* cotton is well documented form all cotton growing countries. The excessive use of insecticides in cotton is due to development of resistance to insect pests due to lack of insecticide resistance management strategy, improper surveillance resulting in delayed application and inadequate control at higher population, excessive use of fertilizer and irrigation water, wrong plant geometry resulting in failure of insecticide to hit the target, resurgence of sucking pests (Dhawan *et al.,* 2000) and lack of proper spray technology. This resulted in loss of synthetic pyrethroids -good molecules for cotton pest management due to development of resistance and resurgence of sucking pests. Similarly insecticides of carbamate (carbaryl) and organophosphates (dimethoate, formothion, phospahmidon, fenitrothion, monocrotophos, profenophos, chlorpyriphos, triazophos, ethion, quinalphos, acephate etc) lost efficacy against key pests of cotton. New chemistry introduced after introduction of *Bt* cotton also faced the same problems, chloronicotnyl provided effective control of sucking but within span of 10 years has developed resistance to cotton jassid which is key sucking pest. Similarly, other sucking pests have developed varying level of resistance to different insect pests. Failure of cotton crop in north irrigated *hirsutum* cotton in north India indicate collapse of insecticides based pest management in cotton.

CONCLUSIONS

Management of cotton pests resulted in development of the concept of Integrated Pest Management. However, for the last five decades the IPM in cotton is dominated by use of insecticides. Unfortunately the use of insecticides in field is not based on proper surveillance. even in developed countries. Non chemical approaches are recommended to manage the pest, but adoption is low due to various constraints. Introduction of *Bt* cotton has reduced the sprays against bollworms but damage of non lepidopteran insects has increased. The introduction of *Bt* cotton susceptible to sucking pests and wrong plant type by private sector may will favour the build of minor pests which will increase pest problems and the use of insecticides leading to over exploitation which may result in failure of insecticide based pest management. For judicious use of insecticides with introduction of area specific new varieties suitable for agro climatic zones and crop protection with suitable spray technology are key issue. The coordination of policy planners, scientific institution and extension agencies is essential to avoid the excessive use of insecticides.

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Population dynamics of whitefly influenced by weather parameters and its management in cotton

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Agriculture continued to change as per need of the society, leading to intensification in crop production practices. India is primarily an agrarian state, nearly 50 percent of the population directly or indirectly earn from agriculture. Farm sector generate a sixth of country's GDP and 14 to 16% contribution is from cotton. Natural fiber has its own importance in the welfare of human beings and main sources are cotton, jute, sheep and silkworm. But 90 per cent of natural fiber is obtained from cotton crop. The crop is the backbone of textile industry. In fact it is the main source of employment to farmers, farm laborers and textile industry dependent laborers. Foreign exchange is also earned to the tune of 30 per cent. Cotton cultivation in India has tremendous scope as its consumption rises by 3 per cent per annum. Consumption in India is increasing because of population blooming and improvement in economic status of the people.

Pest spectrum of cotton is quite complex and as many as 1326 species of insects have been recorded in the world over (Hargreaves, 1984) but in India, 162 species have been observed to damage cotton crop. Out of these, 10 have attained the status of pests and eight are key pests (Dhawan, 2000). Due to insect infestation both quality and quantity is greatly hampered. Premature boll opening results into lint damage and discoloration, immature fiber takes dye differently, lacks durability, forms neps, breakage occurs during spinning and woven fabric is uneven in color and texture.

Since the turn of the 20th century, the quest for the solutions to pest problems has been a dominating concern for this high-valued crop. Perhaps more than any other crop, cotton has been central to the development of integrated pest management as a science and a philosophy. Secondary pest outbreaks, insect resurgence and insecticide resistance have been the central stage problem in cotton cultivation. Even when IPM technology is apparently available, the drive to control insects through a single tactic, such as insecticides, tends to dominate cotton insect control.

Cotton ecosystem : For considering the management practices of any insect-pests, an understanding of the ecological consideration has the paramount importance. Cotton production is a highly complex. Successful insect management makes a difference for farmers. The genetic and environmental factors play an important role in plant growth and development and indicate the periods of plant susceptibility to pests and against insect damage. Cotton field is an integral part of a landscape. Smith and Reynolds (1972) described the major components of the cotton ecosystem which include plants, weeds and other plants, soil and its biota, overall conditioning of the physical and chemical environment, pest species with their natural mortality factors, including parasites, predators and pathogens, arthropod competitors for food and

space, and overall conditioning of humans, including their management of the system.

Charles Darwin (1859) in his classic work on the "Origin of Species" was the first to put forward the idea of environment composed of a number of components which might act separately or jointly to influence the animal's chance to survive and multiply. This idea is well accepted. The biotic and abiotic factors determine the distribution of species in time and space (Yazdani and Agarwal, 1997). The impact of abiotic factors, particularly, climate and their influence on reproduction, development and survival of insects at the individual and population level has become fairly established. However, the impact of various factors interacting with each other remained an unsolved problem to ecologist for a long time.

Significance of whitefly in cotton production: The whitefly infestation remains in cotton more or less throughout the cotton season but the maximum damage in cotton is done during August-September. Higher population has been noticed in dry weather conditions which encourage its population build up. Average six to eight nymphs per leaf is considered its economic threshold.. Using of large amounts of broad-spectrum chemical insecticides to control the key pests of cotton not only creates health problems and environmental pollution, but also develops insecticide resistance in insects (Mohyuddin *et al.*, 1997). Broad-spectrum insecticides use has been reduced to a greater extent with the introduction of *Bt* cotton but consequently with piercing-sucking mouth-parts, whiteflies, survive better (Xu *et al.*, 2008) and feed on their host more comfortably. Whitefly, *Bemisia. tabaci* (Genn.) is a key pest of many field and horticultural crops, throughout the subtropical region (Naranjo, 2001; Bayhan *et al.*, 2006). It damages the host plants directly by depriving them of their nutrients by continuously sucking the cell sap. As this insect-pest acts as vector and helps in the transmission of the viral cotton leaf curl virus diseases (Malik *et al.*, 1999).

Influence of weather parameters on whitefly population: The population dynamics studies of whitefly under the changed agro-climatic conditions are essential to evolve the sustainable pest management strategies. The period of activity of whiteflies lasts from the emergence of seedling to the full grown crop. During the end of cotton season the adults migrate to other crops such as crucifers, cucurbits and malvaceous plants. They migrate to new season cotton crop as soon as it is in the field. High temperature and scanty rainfall situations aggravate the severity of the pest.

Weather factors played an important role in population dynamics of sucking pest insects. The weather factors affect the whitefly population differently in different geographical locations. Sharma and Kumar (2014) proposed that that maximum temp, minimum temp, evening relative humidity and rainfall reduce whitefly population and morning relative humidity and sunshine hours (SSH) are helpful in population build up. Sharma *et al.,* (2004) held the notion that the population dynamics of whitefly on cotton had a non-significant negative correlation with rainfall and relative humidity and positive correlation with temp. However, Ashfaq *et al.*, (2011) found temperature helpful in whitefly population build up and rainfall other way round. On the basis of a case study by Akram *et al.*, (2013) in Multan, Pakistan, concluded that *Bt* genotypes are more susceptible for the whitefly than non-*Bt* genotypes. Regression models for whitefly revealed that minimum

temperature was the most important factor (*Bt* and non *Bt* varieties). Maximum temperature was the major contributing factor for whitefly fluctuation on *Bt* varieties. Jeyakumar *et al.* (2008) noticed higher incidence of whitefly in *Bt* cotton hybrids than non *Bt* cotton. The situation might be due to limited or no feeding by bollworms and not because of higher whitefly susceptibility (Wilson *et al.*, 1992). Among all the abiotic factors, the most important abiotic factor is temperature which has dominant role in pest population variation (Bale *et al.*, 2002). According to Pedigo, (2002) these parameters affects egg laying, increase rate of feeding, metabolism, herbivory and development.

Management: In cotton pest management, strategies have to face with complex of insect-pests. So, the choice of tactics will depend upon the pests and its importance. Sucking pests during early phase of crop growth are the key pests and their control is essential for good production of cotton crop. Eco based IPM is an essential component for a sustainable cotton production. It comprises a series of measures which help in keeping the insect pests below economic threshold. Such control methods include natural control agents, host plant resistance, manipulation of agronomic factors such as rotations, spacings, time of sowing and fertilizer applications besides biological control and use of botanicals.

Cultural practices:

Cotton whitefly and leaf curl virus disease management : In initiation of the cultural practices for whitefly management, the first step kept in mind is early sowing of the crop along with recommended spacing and fertilizers. Late sowing of the crop considerably reduced the yields. Cultivation of susceptible varieties in the established endemic area be immediately discouraged. The movement of diseased plants and its parts must be restricted implementing quarantine measures. The population of the whitefly can be checked on trap crops and inter crops. Alternate weed hosts of *B. tabaci* should be removed from the field. Growing of okra, solanaceous, cucurbitaceous and other alternate host crop in cotton growing tracts should be avoided. Near citrus orchards, cotton crop should not be grown. Desi cotton varieties are resistant to leaf curl disease. In hot spot/ endemic areas, prefer growing of desi cotton. Excessive use of nitrogenous fertilizers should be avoided because the excessive use of nitrogenous fertilizers makes the crop more succulent and susceptible to the pests and disease and ultimately reduced resistance. Install yellow sticky traps for mass trapping of whitefly populations. Seed treatment with Acetamiprid 30-40 g a.i./ ha, Thiomethoxam 25 g a.i./ ha, Imidacloprid 25 g a.i./ ha. NSKE 5 per cent or neem formulations (1500 ppm) 2.5 l/ ha and spraying of Triazophos 40EC 1.5 l/ ha. Using of synthetic pyrethroids should be avoided as they enhance the fecundity in whitefly and cause resurgence. Spraying of the insecticides should be thoroughly cover the lower surface of the cotton leaves for effective control of whitefly.

Biochemical factors and host plant resistance in cotton : The biochemical factors include the allelochemicals and nutrional components in host plant resistance. The plant defense mechanisms involve an elaborative array of phytochemicals and there qualitative and quantitative composition in cotton plant which have the adverse effect on the physiological process of insects after ingestion and can be exploited for cotton pest management as antibiosis (Table 1). The allelochemicals

Table 1: Resistance potential of various biochemical plant traits against different insect pests of cotton

Plant traits	ABW bollworm	SBW bollworm	PBW	Jassid	Thrips	Aphid	Whitefly	Spider mite
High gossypol	R	R	R	R	-	-	-	-
Heliocides(H1 to H4) HH4)	R	R	-	-	-	-	R	-
Silicated leaf	-	-	-	R	-	-	-	-
High tannins	R	R	R	-	-	-	R	R
High bud source	S	-	S	-	-	-	-	-

R= Relative resistance, S= Relative susceptibility, E= Escape
Source: Ilango and Uthamasamy (1989)

compounds known to exert adverse effects on pests in cotton include gossypol, gossypurinn, heliocides and related terpenoids, p-hemigossypolone, tannins, anthocyanins, flavonoids and phenolics.

Tannin is considered to be contributing towards the resistance of plants against insect-pests build up. Strong negative correlation (r = 0.7543) between the tannin content and the cotton whitefly eggs was obtained. The studies highlighted the role of tannins in cotton for imparting resistance against whitefly. Phenolic compounds have been reported to have significant effect on the abundance of several insect species, e.g. total phenols show negative correlation with whitefly population. Total phenols, flavonols, gossypol and ortho-dihydroxyphenols were found associated with the reduction of whitely.

Nutritional factors and host plant resistances : The host plant may be deficient in certain nutritional elements required by the insect and hence prove resistant. The nutritionally deficient plant may cause antibiotic and antixenotic effects on the insects. The antibiosis may result from the absence of certain nutritional substances in the host plant, deficiency of nutritional materials and /or imbalance of available nutrients (Table 2).

Continuous efforts have been made to evolve genotypes with in-built resistance due to nutritional factors for whitefly, *Bemisia tabaci*. Sugar contents in plans support the whitefly increase. Total sugar content of cotton cultivars was positively correlated with whitefly incidence during the vegetative phase but negatively correlated with it during reproductive phase of the crop but Rao *et al.*, (1996) studied the relationship of biochemical and nutritional components of cotton (*Gossypium hirsutum*) leaf with the incidence of whitefly, *B. tabaci*. The resistant genotypes showed higher content of K, P and Mg and lower of N, Fe as compared to susceptible ones.

Table 2. Host plant resistance to whitefly due to nutritional factor in cotton

Insect Pest	Nutritional Factors	Effects	References
Bemisia tabaci (Gennadius)	'!K, P, Mg and"!N, Fe"!Total sugars and non-reducing sugars'!Total sugars	'!Resistance'!Susceptibility (during Vegetative phase)	Rao *et al.,* (1996)
		'!Susceptibility (during reproductive phase)	Rao *et al.,* (1996)
			Butter *et al.,* (1992)
			Butter *et al.,* (1992)

Biological control : Natural enemies (parasitoids, predators, pathogens etc.) contribute to substantial decrease in pest populations where there is least use of chemical insecticides. Naturally occurring native predators *viz.*, *Chilomenes sexmaculatus* and *Chrysoperla zastrowi sillemi* offer significant control of the early season sucking pests. The use of bio-pesticides, *viz.*, *Bt* and viral formulations, assume significance in the management of pests and avoiding the development of resistance.

Parasitoids:

Parasitoids	Pest	Stage of pest
Encarsia formosa	Whitefly	Nymph
Encarsia shafeei	Whitefly	Nymph
Eratomocerus mundus	Whitefly	Nymph

Predators:

Predators	Pest	Stage of pest
Brumoides suturalis	Whitefly	Nymph
Coccinella septumpunctata	Sucking pests	Nymph, Adult, Egg
Chrysoperla carnea	Sucking pests	Egg, Nymph, Adults
Menochilus sexaculatus	Sucking pests	Egg, Nymph, Adults
Scymnus seriacus	Whitefly	Nymph, Adult, Egg
Verania vincta	Whitefly	Nymph

Pathogens:

Pathogen	Pest	Stage of pest
Aspergillus sp.	Whitefly	Nymph

Botanicals : Use of various botanicals against insect-pests are known source of imparting resistance as well as eco-friendly. Neem seed kernel extract @ 5 per cent, neem formulations @ 2 l/ha and neem or karanj oil @ 1%, having antifeedent / deterrent properties are recommended against sucking pests. Neem oil, neem seed kernel extracts and many commercial formulations were found effective for whitefly management. Verma *et al.* (1989) found neem oil (3 l/ha) in combination with Teepol in ratio of 5:1 98.76 per cent control of whitefly against 96.72, 96.52 and 66.47 per cent in quinalphos, amitraz and trizophos, respectively., In Tamil Nadu neem oil (0.5%) was found effective in managing the whitefly population (Jayaraj *et al.*, 1986). Natarajan and Sundramurthy (1990) also observed neem oil to be very effective against nymphs of whitefly. The treated nymphs showed exudates of body fluid 2 days after spray and severely affected nymphs died within three days.

Chemical control : Whitefly management should be achieved when their populations are at low levels through cultural practices. For effective management of the pest, crop ecosystems must be least disturbed. Maintenance of good field sanitation by destroying and removing the crop residues, and weeds is an effective practice against whiteflies. Growing vegetables in short periods and allowing maximum time between host crops of whitefly reduces its pest status on cotton. Combination of cultural practices and need based insecticidal applications keep a check of whitefly populations (Vennila and Biradar, 2007). Repeated applications of insecticides during early and mid seasons lead to resurgence of whiteflies, and hence a highly judicious chemical application is a must. Never use Fipronil, synthetic pyrethroids or any insecticide mixtures. Application of most of the contact and systemic insecticides (monocrtoophos, acephate etc) were observed to increase whitefly build up. Neem oil (1%), fish oil resin soap (2.5%) and neem seed kernel extract (NSKE) 5 per cent give effective control of whiteflies. Triazophos 40 EC @ 600 g a.i./ha, Ethion 50 EC @ 1000 g a.i./ha and Acetamiprid 20 SP @ 30-40 g/ha are effective against whiteflies (Vennila and Biradar, 2007).

More than 25% of leaf coverage by the whitefly pupae on the under surface of leaves of middle plant canopy and flight of white adults visible on a single stroke of the plants should be used to decide the insecticidal applications. The severity of whiteflies is seen after the crop growth crosses 10 nodes on the main stem. Therefore, the amount of spray fluid while spraying the insecticides should be greater than 250 l/ha using power sprayers. Proper coverage of underside of leaves during the insecticidal sprays effectively reduces the whitefly population. Spraying of methyl demeton 25EC @ 500-750 ml/ ha, *Neem* oil plus teepol @ 3-3.5 + 500 ml/ ha, fish oil resin soap @ 14-15 kg/ ha and phosalone 35EC @ 2.5-3 l/ ha. If needed insect growth regulators such as difenthiauron, buprofezin, spiromesifen, and pyriproxifen can be used after mid August. These insecticides are effective on whiteflies and are relatively safer to its natural enemies.

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Increasing trend of biopesticides in the cotton production technologies

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Two thirds of today's world population depends upon agriculture for livelihood, but nowadays, growth and production of agricultural crops is getting hampered day by day due to insect pests and diseases (Elumalai and Rengasamy, 2012). When farmers see their agricultural crops declining in yield and production, they often expect a dramatic, magical treatment to make them lush, green, and healthy again, so that the productivity increases. As a result, they start using chemical pesticides, disregarding their future effects. The extensive use of these synthetic organic chemicals in the past decades has led to a number of long-term environmental problems (Arora *et al.,* 2012). Keeping all these facts in mind, a very big challenge in the new millennium is to produce more and more food from shrinking per capita arable land, keeping the environment safe and innocuous. By the advent of greener approach of developing and using bio pesticides, the situation is gradually changing but in fact can move far more swiftly in this direction which will be sustainable and eco friendly (Mishra *et al.,* 2015). Although bio-pesticides are slowly replacing the chemical pesticides, a complete global look at the scenario indicates that the farmer and particularly the industries based on them are still in an insecure position in comparison to the chemicals which rule the agriculture.

Scenario of cotton production in India : Over the years, country has achieved significant quantitative increase in cotton production. Till 1970s, country used to import massive quantities of cotton in the range of 8.00 to 9.00 lakh bales/annum. However, after Government launched special schemes like intensive cotton production programmes through successive five-year plans, cotton production received the necessary impetus through increase in area and sowing of hybrid varieties around mid 70s (Rajendran and Jain, 2004). Since then, the country has become self-sufficient in cotton production barring few years in the late 90s and early 20s when large quantities of cotton had to be imported due to lower crop production and increasing cotton requirements of the domestic textile industry (http://cotcorp.gov.in/national-cotton.aspx).

Since launch of **"Technology Mission on Cotton"** by Government of India in February 2000, significant achievements have been made in increasing yield and production through development of high yielding varieties, appropriate transfer of technology, better farm management practices, increased area under cultivation of *Bt* cotton hybrids etc. Introduction of *Bt* cotton has played a pivotal role which brought down more than 50 per cent insecticide usage on cotton and 30 - 40 per cent increase in

productivity due to effective pest control and reduction in crop damage (Kranthi, 2012).

Shift of wheel from pesticides to bio pesticides : It is well known that there have been some discoveries in the past which not only have changed the life of man but also had major influence on the globe, and a very well-known chemical pesticide para dichloro diphenyl trichloroethane (DDT) was one of them (West and Campbell, 1946). There are myriads of incidences dealing with DDT poisoning that are already known and some are needed to be further explored (Hill and Robinson, 1945; Dresdend, 1948; Keane, 1972; Tschirley, 1973; Longnecker *et al.,* 1997; Conis, 2010 and Qiu, 2013). DDT was not the only culprit; other categories of synthetic pesticides such as organophosphates (OP), carbamates and pyrethroids were also launched, and by 1980, their impact on pest control and environment was also well recognized (Aktar *et al.*, 2009).

In the eighteenth century and even in the beginning of the nineteenth century, the focus of biological control was to use animals such as birds and entomophagous insects; microbes were not even properly known at that time. The discovery of *Bacillus thuringiensis* (*Bt*) showed a wider aspect of microbe-based biological control (Aronson *et al.,* 1986; Martin and Traverse, 1989; Siegel and Shadduck, 1990; Marrone, 1994; Joung and Cote, 2000). Microbial pest control was a very new concept, and its selective action on pest attracted the concentration of researchers and industrialists equally, and soon the first commercial *Bt* product, Thuricide, was registered in the USA in 1961 (USEPA, 1998). Since then, different sub-species, varieties, and strains of *Bt* have been identified that are effective against a variety of insects (Gonzalez *et al.,* 1982 and Carlton, 1988). In a span of very few years, *Bt* has covered up to 90 per cent of the whole bio pesticide market (Chapple *et al.,* 2000 ; Chattopadhyay *et al.,* 2004 and Romeis *et al.,* 2006), and several *Bt* strains are now registered as bio-pesticides throughout the world (Glare and Callaghan, 2000).

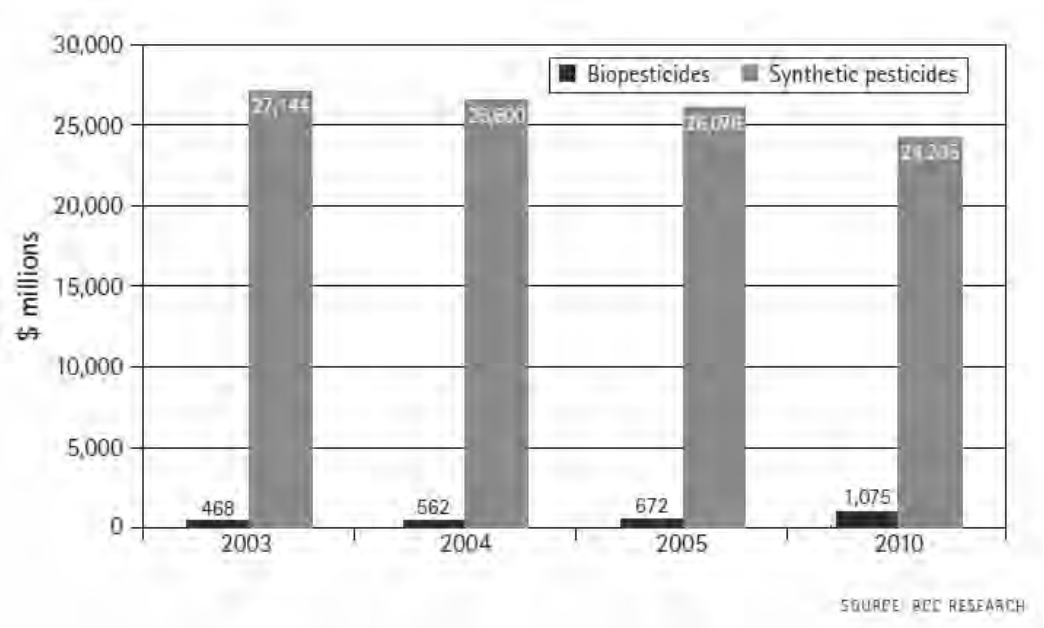

Fig. Global bio pesticides and synthetic pesticides market, 2003–2010

Bio pesticides in world and India : With the increased environmental awareness and the pollution potential and health hazards from many of the conventional pesticides, the demand for nature-based bio-pesticides has been increasing steadily worldwide. The current global organically cultivated agricultural product market is valued at approximately $28 billion. Globally, about 22 million hectares are now organically cultivated. This represents less than 1 per cent of the world's conventional agriculture production and, therefore, a tremendous growth potential for the use of bio-pesticides (Yatin, 2006).

India has vast potential for bio pesticides. However, its adoption by farmers in India needs education for maximizing gains. Bio pesticides represent only 3.59 per cent (as on 2010-11) of the overall pesticide market in India. In India, so far only 12 types of bio-pesticides have been registered under the Insecticide Act, 1968. Neem based pesticides, *Bacillus thuringensis*, NPV and *Trichoderma* are the major bio-pesticides produced and used in India. Whereas more than 190 synthetics are registered for use as chemical pesticides. As of September 2015, there are 436 registered bio pesticide active ingredients and 1401 active bio-pesticide product registrations (http://www2.epa.gov/). Most of the bio pesticides find use in public health, except a few that are used in agriculture wherein, transgenic plants and beneficial organisms are used for pest management in India.

Classes of bio pesticides : Bio-pesticides are certain types of pesticides derived from such natural materials as animals, plants, bacteria, and certain minerals. They include a broad array of microbial pesticides, bio chemicals derived from micro-organisms and other natural sources, and processes involving the genetic modification of plants to express genes encoding insecticidal toxins (Salma *et al.,* 2011).

Bio pesticides fall into three major categories

1. Biochemical pesticides: Naturally occurring substances that control pests by non-toxic mechanisms. Conventional pesticides, by contrast, are generally synthetic materials that directly kill or inactivate the pest. Biochemical pesticides include substances that interfere with mating, such as insect sex pheromones, as well as various scented plant extracts that attract insect pests to traps. In 1979, the U.S. EPA registered the first insect pheromone for use in mass trapping of Japanese beetles. In the 1990s, researchers began testing kaolin clay as an insect repellent in organic fruit orchards. It was made commercially available, particularly for use in organic systems, in 1999.

Spinosad 48 SC, a natural insecticide is a mixture of two derived from fermentation technology produced by *Saccharopolyspora spinosa,* a new species of actinomycete. Spinosad 48 SC is selectively active on insect pests belonging to Lepidoptera. An experiment was conducted to evaluate the bio efficacy of Spinosad 48 SC at four dosages. *Viz.,* 25, 50. 75 and 100 g a.i./ha for two seasons. Results indicated that Spinosad 48 SC at higher dosage levels recorded lower leafhopper and aphid population with average whitefly population reduction. Spinosad 48 SC @ 100 g a.i./ ha recorded minimum per cent bollworm incidence and was *on par* with its lower dosage 75 g a.i. /ha treatment. Maximum good opened bolls and minimum bad opened bolls/plant with higher cotton yield was recorded in Spinosad 48 SC at 100 g a.i./ha. Spinosad 48 SC + Chlorpyrifos @ 25+500 g a.i./ ha combination treatment was *on par* in all the parameters to Spinosad 48 SC alone at 100 g a.i./ ha treatment. Spinosad 48 SC has its

combination of excellent contact and residual efficacy on target pests and safety to beneficial insects and low quantity of application is required for effective management of cotton bollworm. *Helicoverpa armigera* (Hubner). Spinosad 48 SC fits very well in cotton IPM system under irrigated conditions (Patil *et al.*, 1999.)

Studies conducted by Sreenivas and Patil (2001) revealed that commercial neem products were screened for two seasons against cotton insect pests. Pooled data of two seasons indicated that two neem products namely Rakshak and Limnol @ 5.0 l/ha were as effective as endosulfan 35 EC @ 1050 g a.i / ha in reducing the cotton bollworms damage, but were not effective against cotton leafhoppers. Further, neem products were fairly safe to *Trichogramma* egg parasitoids. The role of *neem* products' integration with egg parasitoids and insecticides was studied and found to be fitting well in insect pest management in cotton. Similarly, laboratory study was conducted by Gupta and Raghuraman (2004) using a control group consisting of *Helicoverpa armigera* that was reared on artificial diet without any *neem* treatment. Different concentrations of *neem* (*Azadirachta indica*) (1500 ppm Godrej Achook, 50 000 ppm Neemazal and 60000 ppm *Neem Jeevan* Triguard) were prepared by serial dilution method and their bioefficacy was studied by an artificial-diet surface incorporation technique. The formulations containing azadirachtin-rich content were highly toxic to *H. armigera*. Both Neemazal and Neem Jeevan Triguard at 0.15 per cent showed the best antifeedant effects (73 %). The juvenomimetic effects against *H. armigera* were also studied. The developmental time was significantly longer for all the azadirachtin-rich formulations compared to the control. Larvae treated with Neemazal and *Neem Jeevan* Triguard required 35.8 and 11.1 per cent additional days, respectively, to reach the pupal stage compared to the control. Azadirachtin caused deformities in the developing larvae, pupae and adults. The most marked morphogenetic effects that appeared were larval pupal intermediates, deformed pupae, and adults with frizzled or curved wings, weak and smaller in size. At 0.025 and 0.05 per cent azadirachtin, inhibition and disruption of moulting were observed; however, larval-pupal intermediates and abnormal pupae were also commonly observed. The larval-pupal intermediates were observed at moderate concentrations (0.075 % of Neemazal and 0.10 per cent of *Neem Jeevan* Triguard) resulting in 18.7 per cent of the larval pupal intermediates. The pupal period was also prolonged, where pupae required 43.5 per cent of additional days to reach the adult stage in both the formulations compared to the control. Pupal weight was also significantly lower compared to the control. The pupae that survived either failed to develop further or if developed into adults, died during eclosion with frizzled or curled wings.

2. Microbial pesticides: consists of a micro organism (*e.g:* a bacterium, fungus, virus or protozoan) as the active ingredient. Microbial pesticides can control many different kinds of pests, although each separate active ingredient is relatively specific for its target pests. They offer the advantages of higher selectivity and lower or no toxicity in comparison to conventional chemical pesticides (MacGregor, 2006). These include bio fungicides (*Trichoderma*, *Pseudomonas*, *Bacillus*), bio herbicides (*Phytophthora*), and bio insecticides (*Bt*) (Gupta and Dikshit, 2010). The most widely used microbial pesticides for insects are sub-species and strains of *Bacillus thuringiensis*, or *Bt*. Each strain of this bacterium produces a different mix

of proteins and specifically kills one or a few related species of insect larvae. While, some *Bt* ingredients control moth larvae found on plants, other *Bt* ingredients are specific for larvae of flies and mosquitoes.

Experiments involving biological control for insect pests in agriculture date back to 1835, when Agostine Bassi demonstrated that white-muscadine fungus (*Beauveria bassiana*) could be used to cause an infectious disease in silkworm. The first, and still most, widely used biopesticide included spores of the bacteria *Bacillus thuringiensis* (*Bt*). In 1901, *Bt* was isolated from a diseased silkworm by Japanese biologist, Shigetane Ishiwata. Ernst Berliner in Thuringen, Germany, then rediscovered it ten years later in a diseased caterpillar of flour moth. The *Bt* pathogen was classified in 1911 as type species *Bacillus thuringiensis* and remains the most widely used bio pesticide to this day. In the early 1920s, the French began to use *Bt* as a biological insecticide. The first commercially available *Bt* product, Sporeine, appeared in France in 1938. In US, in the 1950s, widespread use of bio pesticides began to take hold as a host of research on *Bt* efficacy was published. In 1973, *Heliothis* NPV was granted exemption from tolerance and the first viral insecticide, Elcar received a label in 1975. In 1977, *Bacillus thuringiensis* var. *israelensis* (toxic to flies) was discovered, and in 1983 the strain *tenebrion is* (toxic to beetles) was found.

Bacterial biopesticides : Bacterial biopesticides are the most common form of microbial pesticides that function in multiple ways. In insects, bacteria disrupt the digestive system by producing endotoxins that are often specific to the particular insect pest (Brien *et al.,* 2009). The members of the genus *Bacillus* are often considered as microbial factories for the production of vast array of biologically active molecules, some of which are potentially inhibitory for fungal growth (Schallmey *et al.*, 2004).

Field study was conducted by Sreenivas and Patil (2001) for two years to evaluate the *Bacillus thuringiensis* commercial products *viz.*, *BT*K 1, *BT*K II, Dipel, Delfin and BARC strain against bollworm. *Helicoverpa armigera* (Hubner) on cotton. The *B. thuringiensis* products were applied four times based on ETL of pest population. Among the various *B. thuringiensis* products, *BT*K II recorded lowest bollworm damage highest GOB, lowest BOB and maximum cotton yield which was *on par* with Dipel 8 L. *Bacillus thuringiensis* products are detrimental to all stages of mulberry silkworm, *Bombyx mori* and mortality was recorded up to 40 days when treated mulberry leaves are fed to worms. Mortality of silkworm was observed in Dipel spray drift at 5 m and 20 m distance with knapsack and power sprayer respectively.

A comparative study of *Bacillus thuringiensis* on cotton bollworms was carried out by Rehman *et al., (*2002). Three *Bacillus thuringiensis* biopesticides were sprayed at their recommended rate against cotton bollworms, which have controlled these pests effectively both in laboratory and field trials. In the lab, CAMB- *Bt* was found the best bio-pesticides giving 100 per cent mortality with in 48 hrs against spotted bollworms. Similarly, Lepinox-*Bt* gave 100 per cent mortality against spotted bollworms. However, it was least effective against American bollworm with less than 60 per cent killing. Comparatively Larvo-*Bt* gave less than 20 per cent mortality against both the insects.

Fungal biopesticides : *Beauveria bassiana* (Balsamo) Vuillemin and *Metarhizium anisopliae* (Metchnikoff) Sorokin are naturally occurring entomopathogenic fungi that infect sucking pests including *Nezara viridula* (L) (green vegetable bug) and *Creontiades* sp. (green and brown mirids) (Gomez and Moscardi, 1998). Fungi have the unique ability to attack insects by penetrating through the cuticle making them ideal for the control of sucking pests. Studies also show that *B. bassiana* is virulent against *Lygus hesperus* Knight (Hemiptera: Miridae), a major pest of alfalfa and cotton in the USA (Noma and Strickler, 2000).

Viral biopesticides : Like bacteria and fungi, viral biopesticides play a significant role in antagonizing pathogens especially bacteria in the form of bacteriophages. Apart from it, viruses are host specific, infecting only one or a few closely related species, thus offering minimal off-target impacts (Cory and Myers, 2003 ; England *et al.,* 2004 ; Raymond *et al.,* 2005 ; Hewson *et al.,* 2011). Baculovirus is the main virus that is commercially used for designing phage pesticides. Since the start of their commercial use, baculoviruses have been tested extensively to assess their safety in order to meet registration requirements (Burges *et al.* 1980; Groner 1986; Ignoffo, 1975). Baculoviruses develop in the nuclei of the host insect cells. When ingested by the host insect, infectious virus particles are liberated internally and become active. Once in the larval gut, the virus's protein overcoat quickly disintegrates, and the viral DNA proceeds to infect digestive cells. Within a few days, the host larvae are unable to digest food and so weaken and die (Yatin, 2006). Baculoviruses are particularly attractive for use as biopesticides due to their high host specificity. Each virus only attacks particular species of insects, and they have been shown to have no negative impacts on plants, mammals, birds, fish, or non target insects (Amico, 2007). Baculoviruses can also cause sudden and severe outbreaks within the host population for complete control of the disease (Sylvar, 2008). Another major advantage of baculoviruses is that in some cases, they can replace and serve as an alternative to the antibiotics and chemical pesticides (Brien *et al.,* 2009).

As of 2010, over 24 baculovirus species have been reported to be registered for use in insect pest management throughout the world (Kabaluk *et al.,* 2010). The market share of baculoviruses is 6 per cent of all microbial pesticides (Quinlan and Gill 2006; Marrone, 2007), and millions of hectares have been treated with registered baculovirus products over the years (Szewczyk *et al.,* 2009; Kabaluk *et al.,* 2010; Moscardi *et al.,* 2011). Despite many years of use and testing against non target organisms, no adverse effects have ever been attributed to baculoviruses (William, 2007).

Sreenivas and Patil (2003) bollworm damage revealed the superiority of HaNPV 500 LE+ edosulfan 35 EC 1050 g a.i application which recorded minimum pest damage and was *on par* with reduced dose of 250 LE + 1050 g. a.i./ha treatment but superior to sole application of HaNPV and untreated control . Maximum number of good opened bolls was registered in HaNPV 500LE + endosulfan 35 EC 1050 g. a.i./ha which was *on par* with all combinations except lowest dosage of HaNPV 250 LE + endosulfan 35 EC 525 g. a.i./ha. Among the adjuvants screened teepol , jaggery and boric acid each at 0.1 per cent proved better for HaNPV as they recorded significant control and higher seed cotton yield.

List of some commonly available biopesticides

Category of biopesticide	Products common name or trade name	Targets
B. popilliae	Milky spore powder	Japanese beetle grubs
Bacillus sphaericus Serotype H5a5bstrain 2362 ATCC1170	VectoLex	Mosquito larvae
B . thuringiensis subsp. aizawai NB200	Florbac	Moth larvae
B. thuringiensis sub sp *israelensis*	BMP	Mosquito and blackflies
B. thuringiensis sub sp *israelensis* EG2215	Gnatrol , Aquabac	flies, Mosquito
B. thuringiensis sub sp *aizawaidelta* endotoxin in killed *P. fl uorescens*	M-Trak	Colorado potato beetle
B. thuringiensis sub sp *aizawai* GC-91	Agree WG	Plutella
B. thuringiensis sub sp *kurstaki*	Thuricide Forestry Wilbur-Ellis *BT* 320DipelDeliverBiobit HPForayJavelin WGGreen LightHi-Yield Worm Spray Ferti-LomeBonideBritz *BT*Worm WhipperSecurity Dipel Dust	Lepidopteran larvae
B. thuringiensis sub sp *kurstaki* BMP 123	BMP123	Lepidopteran larvae
B. thuringiensis sub sp *Tenebrionis*	Novodor	Colorado potato beetle
B. thuringiensis sub sp *kurstaki* EG7826	Lepinox WDG	Lepidopteran larvae
B. bassiana 447	Baits Motel Stay- awhile	Ants
B. bassiana ATCC 74040	Naturalis L	Various insects
B. bassiana GHA	Mycotrol ES Mycotrol OBotanigard 22WPBotaniGard ES	Various insects
B. bassiana HF23	balEnce	Housefl y
M. anisopliae F52	Tick-Ex	Ticks and grubs
Paecilomyces fumosoroseus Apopka 97	PFR-97	Whitefl y and thrips
Nosema locustae	Nolo-Bait Semaspore bait	Grasshopper and crickets
Anagrapha falcifera NPV	CLV-LC	Lepidopteran larvae
Cydia pomonella GV	CYD-X	Virus codling moth
Gypsy moth	NPV	Gypchek Gypsy moth
H. zea NPV (previously *Heliothis zea* NPV)	GemStar	Cotton bollworm, tobacco, budworm
Indian meal moth GV (*P.interpunctella* GV)	Fruitguard	Indian meal moth
Mamestra confi gurata NPV (107308)	Virosoft	Bertha armyworm
Spodoptera exigua NPV	Virus Spod-X	Beet armyworm
Saccharomyces cerevisiae	Bull Run	Fly attractant

A study by Pophum *et al.,* (1997) was conducted on *Helicoverpa zea* Nuclear polyhedrosis virus, which was previously registered and commercially produced as a pesticide (Elcar), was genetically improved to control the cotton bollworm, *H. zea.* A significant reduction in the time required for this virus to kill *H. zea* larvae was achieved by inserting a mite gene encoding a potent insect selective neurotoxin gene, *tox34,* into the viral gene encoding ecdysteroid UDP glucosyltransferase. Under the control of an early viral promoter, expression of *tox34* during infection resulted in 50 per cent mortality or paralysis within 40 h after virus treatment. The ability to genetically improve the properties of this virus as a pesticide provides the opportunity to develop a commercially viable product to control this pest species.

Entomogenous nematodes : An experiment conducted by Sreenivas and Patil (2001) to evaluate the efficacy of commercially available green commondos nematode, *Steinernema feltiae* Filipjev during 1994 and 1995 seasons both in the field and laboratory conditions. Study revealed that the nematodes were applied at 45 lakhs per ha when used alone and 50 per cent together with HaNPV or *Bacillus thuringiensis* or endosulfan 35 EC. Per cent bollworm damage revealed lowest in edosulfan 35 EC treatment which was *on par* with nematodes + half dose of endosulfan 35 EC (16.96%) and combined use of all three pathogens *viz.*, nematodes + HaNPV + *Bt* (@ 45 lakhs + 25 LE + 1.01 /ha). Maximum number of good opened bolls and minimum bad opened bolls with highest seed cotton yield was registered in combined use of all three bioagents treatments which was *on par* with sole application of endosulfan 35 EC. Mortality of *H.armigera* under laboratory conditions revealed superiority of combined use of three pathogens which registered 66.67 per cent mortality of early instars. None of the bioagents registered mortality of late instars up to four days.

3. Plant incorporated protectants (PIPs) are pesticidal substances that plants produce from genetic material that has been added to the plant. For example, scientists can take the gene for the *Bt* pesticidal protein and introduce the gene into the plant's own genetic material. Then the plant, instead of the *Bt* bacterium, manufactures the substance that destroys the pest. The protein and its genetic material, but not the plant itself. The genes that code for the insecticidal crystal proteins have been successfully transferred into different crop plants including cotton, tomato, brinjal, etc. that lead to significant economic benefits. Due to their high specificity and safety in the environment, *B. thuringiensis* and Cry proteins are efficient, safe, and sustainable alternatives to chemical pesticides for the control of insect pests (Roy *et al.,* 2007; Kumar, 2012).

CONCLUSIONS

Developing countries have huge possibilities for using bio-pesticides as the production can be less expensive and labour is cheap in comparison to developed nations (Roettger and Reinhold, 2003). Also countries like India are vastly dependent upon agriculture for not only feeding their populations but also for the economy which depends majorly on this sector. However, most of the challenges faced for the upliftment of bio-pesticides are fundamental and cosmopolitan. These include the efficacy of the microbial activity, survival of microorganisms, delivery systems, determining

host range, and avoiding injury to non-target organisms, consistency, performance in field conditions, economics, government regulations, and confidence among the end users.

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Cotton leaf curl disease (CLCuD) serious threat to cotton in north India - An overview

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Cotton is the most important *kharif* cash crop of north India. Among the various factors responsible for its low production and productivity during the last two decade, cotton leaf curl virus disease (CLCuD) has been found to be one of the major limiting factor. The disease has assumed serious proportions in the most potential irrigated cotton belt of north India comprising an area of around fifteen lakh hectares. The disease caused by a whitefly transmitted Gemini virus was first noticed in Nigeria on *Gossypium peruvianum* and *G. vitifolia* (Farquharson, 1912). In India, cotton leaf curl virus disease was first reported on American cotton (*G hirsutum*) in Sriganganagar area of Rajasthan state during 1993 (Ajmera,1994) and during 1994 it appeared in Haryana and Punjab (Rishi and Chauhan,1994; Singh *et al.,*1994) states on *hirsutum* cotton and posed a major threat to its cultivation in northern India (Varma *et al.,*1995). The disease has appeared in an epidemic form during 1997 in the Rajasthan a and inn Punjab affecting an area of 0.1 million hactares (Anonymous,1998). The major area (more than 90%) has now come under *Bt* cotton hybrids. Cotton leaf curl virus disease appeared in a severe form during 2009-2010 crop season in some areas of north zone (Anonymous, 2011). The hitherto known resistant varieties also showed susceptible reaction at hot spot areas. Recent advances made in development of new resistant varieties/hybrids, epidemiological studies including development of disease maps and detection of new weed hosts and breakdown of resistance due to development of new viral recombinants are discussed along with future management strategies.

A class of *Geminiviruses* was observed in 1978, with distinct characteristics of size and appearance of geminate particles and was subsequently proven to be evidence that this is single-stranded deoxyribonucleic acid (ssDNA) virus (Mathews, 1987). The family *Geminiviridae* comprises of three genera *i.e. Mastrevirus, Curtovirus* and *Begomovirus.* A notorious group of these viruses belongs to genus *Begomovirus*, cause of major threat to cotton crop which is well known as Cotton leaf curl virus disease. *Begomoviruses* have been found associated with the disease in the Indian subcontinent specifically Cotton leaf curl Multan virus (CLCuMV), Cotton leaf curl Rajasthan virus (CLCuRV), Cotton leaf curl Alabad virus (CLCuAV), Cotton leaf curl Kokhran virus (CLCuKV) and Papaya leaf curl virus (PaLCuV), Cotton leaf curl Bangalore virus (CLCuBV) alongwith associated alpha-satellite and beta-satellite molecules (Mansoor *et al.,* 2003; Briddon *et al.,* 2003) and Cotton leaf curl Burewala virus (CLCuBuV) (Amrao *et al.,* 2010). CLCuD is recorded as one of the disparaging diseases of cotton. Cotton leaf curl virus has an attention-grabbing evolutionary story. It was first reported in Nigeria (1912) on *Gossypium peruvianum* and *Gossypium vitifolia*, Sudan (1924), Tanzania (1926), Philippine (1959) but in Pakistan CLCuD

was first recorded in the 1967 in Multan district on scattered *hirsutum* plants (Farquharson, 1912; Hussain and Ali, 1975). It was not well thought-out as a serious disease up to 1987 but appeared in epidemic form in 1992-1993 which caused a decrease in production down to 9.05 million bales and during the 1993-1994 season to 8.04 million bales (Mahmood *et al.,* 2003). The financial losses with the estimated value of $5 billion (US) to the nation occurred from 1992-1997 in Pakistan (Briddon and Markham, 2001). In 1997, CLCuD was reported from Sindh province of Pakistan which was previously free from this disease (Mansoor *et al.,* 1998). It is very complicated to calculate the precise estimates because the incidence of CLCuD varies from year to year and also varies from area to area under cotton cultivation. Cotton belongs to genus *Gossypium*, which comprises of 52 species, of which four are cultivated species including *G. hirsutum* (Allotetraploid), *G.barbadense* (Allotetraploid), *G. arboreum* (Diploid) and *G. herbaceum* (Diploid) (Azhar *et al.,* 2010a). The *hirsutum* species of cotton (Upland or American) are under the attack of Cotton leaf curl disease (CLCuD) since 1970 and ruined the existing variety S-12 and are still slaving the new emerging varieties (Perveen *et al.,* 2010). CLCuD get transmitted by whitefly *i.e. Bemisia tabaci* complex (including *B. argentifolii*) in a persistent manner (Brown *et al.,* 1995; Rybicki and Fauquet, 1998). Most of the *Begomoviruses* comprised of two genomic components called DNA-A and DNA-B, which are indispensable for a disease that is transmitted by whitefly *Bemisia tabaci* (Monga *et al.,* 2011). There are numerous viruses from the Old World which have only a single constituent, analogous to DNA-A, which has been isolated and shown to bring on disease symptoms (Navot *et al.,* 1991; Dry *et al.,* 1993; Tan *et al.,* 1995). The DNA isolated from an infected plant of cotton with CLCuD showed wide-ranging homology with the DNAA component and other *Begomoviruses* in the Indian subcontinent (Zhou *et al.*, 1998).

Appearance and transmission of disease in north India : In Indian subcontinent (Pakistan) the disease was first reported in late 1960s and remained a minor sporadic problem till 1980's. From 1992 to 1997, this disease appeared in very severe form and affected Pakistan economy up to US $ 5.0 billion (Radhakrishnan *et al.,* 2004). The disease was first reported in North India in 1993 from Sriganganagar district of Rajasthan state on *G. hirsutum* (Narula *et al.,* 1999). The first symptoms of disease on cotton in Punjab appeared in 1995 and the disease continued to spread steadily eastwards in Punjab, Rajasthan and Haryana states (Briddon, 2003).The incidence of CLCuD varied from traces to 10; traces to 30 and 10 to 80 per cent in the month of June, July and August respectively on different varieties / hybrids in various cotton growing areas of the Punjab in1998 and 1999 (Singh *et al.,* 2001). From *kharif* 2005 to *kharif* 2010 cotton season, the incidence of CLCuD varied from traces to 8 per cent along with I to III grade severity on different varieties/hybrids *viz.,* F 1861, LHH 144, MRC 6304 *Bt* and F 1378 at different farmers field, research farms of Bathinda, Faridkot, Muktsar and Ferozepur districts on cotton growing areas. Similarily in the 4th. week of July and in 4th. week of August the incidence of CLCuD was traces to 65 per cent and traces to 100 per cent with traces to IV grade severity on varieties/hybrids like F 505, F 846, F 1054, RCH 134 BGII and some undescripts. These findings showed that the incidence/severity of CLCuD increased from June to September in *kharif* 2005, 2006, 2007, 2008, 2009 and 2010 crop seasons in different cotton growing areas of the

state. The maximum incidence and severity of CLCuD was observed in *kharif* 2009 and 2010 on various hybrids /varieties of *American* cotton at farmers field and at research farms. In *kharif* 2010, it appeared in epidemic form in almost all the varieties/hybrids of the cotton. The incidence was traces to 65.0 *per cent* with 0-IIIrd grade symptoms (MRC 7017 and RCH 134 *Bt*); traces to 100.0*per cent* with 0-IVth. grade symptoms (RST 9 and RCH 134 *Bt*) ; traces to 95.0*per cent* with 0- IVth. grade symptoms (F 846 and RCH 134 *Bt*) in Bathinda, Faridkot, Muktsar and Ferozepur area. (Anonymous, 2006-2011). Due to breakdown of resistance from 2011 onwards (*Bt* hybrids like MRC 7017 BGII , MRC 7031 BG II and Anukar 3028 BGII) till date many of the private company other *Bt* hybrids tested in research trials in north zone showed susceptibility/tolerance to CLCuD At present major cotton area under BG II cotton hybrids which are having tolerance and none is resistant to CLCuD in whole of North India. (Anonymous, 2015).

CLCuD causative complex : Most of the begomoviruses comprised of two genomic components called DNA-A and DNA-B, which are indispensable for disease that is transmitted by whitefly *Bemisia tabaci* (Monga *et al.,* 2011). Population build up of whitefly over the years has taken place ut there is no relation of vector population with CLCuD incidence and severity. During 2015-2016, there is whitefly epidemic but leaf curl incidence is traces to low. It is the most vital sucking pest of both industrial and food crops like Cotton, Sunflower, Melon, tomato, Brinjal etc. (Rafiq *et al.,* 2008). Its polyphagous nature is confirmed over 500 plant species all over the world including Asia, Africa, America, Europe, Russia, Australia and Pacific Islands (Greathead, 1986). In cotton growing areas of Central Punjab it has been reported in about 164 plant species (Attique *et al.,* 2003). In 16 of the 27 cotton growing countries whitefly is recognized as a major pest during mid to late sowing time. The variability of DNA beta component of CLCuD causative complex has been established and the sequence has been submitted to NCBI niz., KJ 614434, KJ 614435 and KJ 614436 (Inder *et.al.,* 2014).

Epidemiology of CLCuD : Bink (1975) who reported weather factors viz., temperature, wind and rainfall etc. affected the epidemiology of leaf curl in Africa. In Pakistan the infection of CLCuD in eight of the varieties of *American* cotton was observed at maximum and minimum temperature of 33-45 ^{0}C and 25-30 ^{0}C respectively (Khan *et.al.,*1998). Then Singh *et al.,* (2003) also found that CLCuD percent incidence was significantly correlated with maximum and mean temperature; maximum and mean relative humdity, rainfall and whitefly population in particular seasons. Monga *et al.,* (2004) made prediction equations for the appearance of CLCuD. They found that a maximum temperature between 35-42^{0}C, minimum temperature between 26-29 ^{0}C and maximum relative humidity 71-95per cent in the month of July *in kharif* 2002 and 2003 seasons favoured the maximum development of disease.

Singh *et al.,* (2010) gave prediction of CLCuD in disease prone area using linear model. They reported that mean temperature (30.9-35.2^0 C), minimum relative humidity (47.5 -63.5%) and mean relative humidity (55.5-71.0%) in the month of July favoured the appearance of the disease. They also found that minimum temperature and relative humidity of two lag weeks played very important role in the appearance and progress of the disease. Monga *et al.,* (2011) found that minimum

temperature and sunshine hours showed significant negative correlation, whereas morning relative humidity and rainfall gave positive correlations with incidence and progress of the disease and this regression equation helped in understanding the factors affecting disease development and its prediction. Farooq *et al.,* (2014) studied that climate change and weather fluctuation have profound influence on the spread of Cotton leaf curl virus. They observed that temperature and plant age influenced the Cotton leaf curl virus disease epidemiology.

Host range : Host range that has been identified for CLCuV include *Abutilon theophrasti* (Nill), *Althaea rosea* (Cav.), *A. ficifolia*, *A. kurdica*, *A. nudiflora*, *A. Pontica*, *A. sulphurea*, *G. barbadense*, *G. hirsutum*, *Hibiscus cannabinus* (L.), *H. esculentus* (L.), *H. ficulneus*, *H. huegelii*, *H. trionum*, *H. sabdariffa* (L.), *Lavatera cretica*, *Malva alcea* (L.), *M. silvestris* (L.), *M. moschata* (L.), *Malvaviscus arboreus* Car., *Pavonia hastate* (L.), *Sida acuta* (Burm.), *S. alba* (L.), *S. cordifolia* (L.) and *Nicotiana tabacum* L. (Tarr, 1951, 1957; Bink, 1975; Cauquil and Follin, 1983; Fauquet and Thouvenel, 1987). *G. arboreum* and *G. herbaceum* are resistant to CLCuV (Cauquil and Follin, 1983). Similar to Cotton leaf curl virus symptoms were reported in other plant species in Africa but there is ambiguity whether the same virus is involved in these species or not. These include *Corchorus fascicularis* Lau. (Tilliaaceae), *Phyllanthus niruri* L. (Euphorbiaceae), *Clitoria ternatea* 1., (Fabaceae), *Phaseolus vulgaris* (Fabaceae), *Sida urens* (Malvaceae), Petunia sp. (Solanaceae) and *Urena lobata* (Tarr, 1951; Nour and Nour, 1964; El- Nur and Abu Salih, 1970). In Pakistan under field conditions CLCuV symptoms were observed on alternate hosts like Brinjal, Cucurbits, *Convolvulus arvensis*, *Rumex dentatus*, Water Melon, Cow Pea and Lilly plants (Anonymous, 1993).

Effect of CLCuD on yield and fiber traits : Losses due to CLCuD are dependant on infectivity time and variety. The pronounced damage of CLCuD is at early stage of infection but at later stages results minor infections (Akhtar *et al.*, 2003). CLCuD damage differs on various plant parts and ultimately results in reduction of yield. It can reduce boll weight 33.8per cent, 73.5per cent in bolls per plant, GOTper cent upto 3.93per cent, seed index 17.0per cent and yield/plant 64.5per cent (Ahmed, 1999). The cotton fiber (lint) is the most important commodity for textile industry and CLCuD also affects fiber quality traits (Kalhoro *et al.*, 2002). According to Ahmed *et al.*, (1999) CLCuD can decrease fiber length 3.44per cent, fiber strength 10per cent and elongation percentage upto 10per cent. Akhtar *et al.,* (2009) studied impact of CLCuD on fiber quality traits and the findings depicts that the CLCuD significantly affect traits like GOT, fiber length, fiber uniformity index, short fiber index, fiber fineness, fiber bundle strength yellowness and maturity ratio. In their studies they observed significant affects of this viral disease on cellulose, protein, wax and pectin which are the major constituent of fiber. But in view of Idris (1990) virus has significant impact on yield but not on fiber quality. Singh *et al.,* (2002) reported that CLCuD caused 64.7per cent reeuction in number of bolls along with 49.6per cent reduction in boll weight in susceptible variety F 846. They also found that this disease remarkably deteriorated the quality of fibre *i.e.* fibre length reduced by 2.9per cent, elongation by 13.0per cent, uniformity by 1.6per cent and micrinarire value by 6.3per cent in diseased plants having grade 1, 2, 3 and 4 symptom of disease over 0

grade (healthy plants). Further Singh (2006) found there was 50.4per cent reduction in number of bolls and 42.9per cent in boll weight of plants due to CLCuD infection in variet F 846. This malady reduced the fibre length by 5.2per cent, strength by 5.4per cent , elongation by 10.0per cent, uniformity by 2.2per cent and miconarie value by 4.1per cent in diseased plants over the grade 0 (healthy) plants in variety F 846. In another study Singh *et al.,* (2013) reported that the disease caused 52.7per cent reduction in number of bolls and 54.2per cent in boll weight in *Bt* cotton hybrid RCH 134. Similarly it reduced the fibre length from 29.1 to 26.2 mm (9.9%); fibre uniformity from 68.9 to 68.1per cent (1.1%); fibre strength from 29.1 to 26.9 g/ texture 97.5%) micronaire value from 5.2 to 5.0 g inch^{-1} (3.8%). Similar results were reported in *Bt* cotton hybrid MRC 6304. Production losses due to CLCuD during decades are given in (Table 1).

Overall cotton loss due to major disease CLCuD was in the range of 1.90 to 34.8per cent during the period under study. The overall crop loss due to CLCuD during first decade *i.e.* 2002-2011 was 9.6per cent.

Inheritance of cotton leaf curl disease (CLCuD) : The inheritance of cotton leaf curl disease is still a dilemma among the plant researchers and no comprehensive assessment found about the resistance inheritance of this disease (Khan *et al.,* 2007). The viral resistance in cotton may be an unstable character reported by Tarr (1951). The breeding for cotton leaf curl disease (CLCuD) resistance has been achieved through the assemblage of minor genes by recurrent selection (Hutchinson and Knight, 1950) and according to Azhar *et al.,* (2010b) resistance depends on major genes (dominant genes) which may lose quickly because of the evolution of pathogen for these genes. An alternative approach is needed for partial resistance that depends on the recombination of minor genes (recessive genes). The concept of polygenic mode of inheritance of cotton leaf curl disease was changed into single dominant gene (with minor modifier genes) as determined by Saddig (1968) and also clarified by Ahuja *et al.,* (2006). The cross between *Gossypium barbadense* L. (Giza-45) and *Gossypium hirsutum* L. (Reba P 288) determined the effects of a single dominant gene supported by Aslam *et al.,* (2000). The F1 of crosses between highly susceptible S12, highly resistant LRA 5166 varieties were found having all virus free plants and their F2 was close to 1:3 ratios which exhibit the presence of a single gene for the inheritance of resistance against CLCuD reported by Mehmood (2004) and Rehman *et al.,* (2005), Whereas in the same cross (LRA 5166 × S 12) no single gene of major effect was found to be responsible for cotton leaf curl disease (Khan *et al.,* 2007). Whereas the nuclear inheritance is under discussion, the extra chromosomal inheritance is also a secret and considered to be absent by Rehman *et al.,* (2005) but the presence of reciprocal differences in the cross LRA 5166 is advocated by Khan *et al.,* (2007). Pathak *et al.,* (2009) reported that the resistance to CLCuD is controlled by dominant gene and suggested that cytoplasm of the parental lines does not have any contribution in the inheritance of this disease.

Screening of materials for resistance:

(a) Non *Bt* cultivars / hybrids: Tthe most commonly grown cultivars in the northern states during 1993 when the disease appeared were RST 9 in Rajasthan , F 846 in Punjab and HS 6 in Haryana proved highly susceptible to this

disease. From 1996 till dates several resistant/ tolerant culivars namely RS 810, RS 875, RS 2013 ; LH 1556, F 1861, LH 2076, LH 2108, F 2228 ; H 1117, ,H 1226 and hybrids like LHH 144, CSHH 198, CSHH 238 and CSHH 243 were developed bv RAU, Rajasthan, PAU, Ludhiana, HAU, Hisar and CICR, Sirsa respectively.
(Anonymous, 2005-2015).

(b) BGI and BG II hybrids: From 2005 to 2014 large number of private company tested in research trials in north zone werefound susceptible to CLCuD. At present few hybrids having some tolerance were recommended for cultivation in north zone (Anonymous, 2005-2015).

Breakdown of resistance against CLCuD : The secret of inheritance is still under discussion and another idea of resistance breakdown was initiated that so called achieved resistance has been broken down by virus mutation whereas the symptoms and parameters for identification are still same in practice. García and McDonald (2003) reported that the virus mutation requires 25 years at least then who does it possible after 1967s our researchers get early resistance and instantly destroyed the integrity. Whereas the changing climate scenario, cotton varieties either susceptible are sown early can escape from virus and whitefly but the resistant one could be susceptible in late sowing which is the cause of ambiguity between susceptible and resistant. The concept of polygenic mode of inheritance of cotton leaf curl disease was changed into single dominant gene (with minor modifier genes) as determined by Saddig (1968) and also clarified by Ahuja *et al.,* (2006). The cross between *Gossypium barbadense* L. (Giza 45) and *Gossypium hirsutum* L. (Reba P 288) determined the effects of single dominant gene supported by Aslam *et al.,* (2000). The F1 of crosses between highly susceptible S-12, highly resistant LRA 5166 varieties were found all virus free plants and their F2 was close to 1:3 ratios which exhibit the presence of single gene for the inheritance of resistance against CLCuD reported by Rehman *et al.,* (2005), Whereas in same cross (LRA 5166 × S 12) no single gene of major effect found to be responsible for cotton leaf curl disease (Khan *et al.*, 2007). Then Bhatoa *et al.,* (2009) reported that all the resistant germplasm available in north zone screened at Abohar (hot spot of CLCuD) showed disease upto varying extent. Due to reports of breakdown of resistance in popular cotton hybrids in north India in 2010, six strains of CLCuD , including Sriganganagar strain isolated from a severely infected CLCuD resistant cultivar were characterized and nucleotide sequences of DNA- A and beta DNA components were determined. Results indicated that recombination in several regions of DNA-A and beta DNA in the potential resistance breaking Sriganganagar strain of CLCUD was mapped to the highly virulent Burewala strain and several other strains (Chakrabarty *et al.,* 2011). Further Rajgopalan *et. al.,* (2012) found that the resistance breaking in cotton leaf curl Burewala virus (CLCuBuV) is now dominant virus in many fields in cotton growing areas of Punjab , Haryana and Rajasthan.

Sources of resistance : Number of resistant lines against CLCuD have reported from Sriganganagar, HAU, Hisar, CICR, Sirsa and PAU, Ludhiana (Ajmera *et al.,* 2004; Radhakrishnan *et al.,* 2004 ; Beniwal and Siwach, 2011 ; Bhatoa and Sekhon, 2012). But due to appearance of recombinant strains no

lines out of earlier available lines as well as more than 5000 lines screened during 2011-2015 have remained completely free from disease (Monga, 2014). Thus exploring of *Gossypium thurberri, G. anomalum, G. raimondii, G. armourianum* and *G. tomentosum* are the best sources for resistance against CLCuD (Azhar *et al.*, 2010b). *Gossypium arboreum* , known as desi cotton , is closest species to the 'A' genome donor of the tetraploid cottons and has been observed to be immune to CLCuD (Gupta *et al.,* 2006). Similarly , *G. armourianum* (DD) another related D genome species has been found to be highly resistant to this disease. Despite the evolution of new races of virus and knocking out of established resistant/ tolerant genetic stocks of American cotton , these related diploid cottons have maintained their resistant response to CLCuD. Similarly , resistant triploid interspecific hybrid between *G. hirsutum* x *G. armourianum* is another source of resistance.

Screening methods employed to develop CLCuD tolerant materials artificial screening trough viruliferous whiteflies inoculation : Screening methods those commonly used is the exploitation of virus spreader line (S 12) and white fly as a source of transmission vector (Shah *et al.,* 2004; Perveen *et al.,* 2005). The whitefly required acquisition threshold period of 20 minutes, inoculation access of 10 minutes and a latent period of 8 hours for successful transmission of the virus. Percent transmission increased with increase in acquisition and inoculation access periods (Mann and Singh, 2004). For transmission through spreader line these researcher used S 12 the popular and most susceptible variety to CLCuD disease. This variety was planted in rows among the tested genotypes for natural spreader of disease. Shah *et al., (*2004) proposed whitefly mediated transmission using insect proof cages. Another method that was used for screening is the sowing time difference *i.e.* normal and late sowing along with disease nursery (Ahuja *et al.*, 2006; Perveen *et al.*, 2010). They established CLCuD nursery near the experimental area to allow the spread of whitefly vector throughout the season and tested different sowing dates.

Another method to screen the germplasm against cotton leaf curl virus is through inoculation using veruliferous whiteflies in net house conditions either by open choice method or through release of counted veruliferous flies on test plants under plastic jars in polyhouse for fixed interval (Monga *et al.*, 2011)

Through grafting : Grafting is the most efficient method to transmit the causal agent as grafted plants develop symptoms within 14-30 days depending upon varietal susceptibility/ resistance (Akhtar *et al,.* 2001; 2002b). Grafting as a successful method to inoculate CLCuD was used by (Akhtar *et. al.,* 2002 (a, b, c), 2004, 2010; Shah *et al.,* 2004). For grafting researchers employed three procedures like bottle graft, top cleft and wedge graft. In this procedure the stock used as resistant and scion as susceptible source for inoculation of disease and later presence of virus was confirmed by the use of ELISA.

PCR can be used as a reliable tool for the detection of viruses. As the geminiviruses are small, single stranded and have circular genome thus PCR can be efficiently used for their detection. Several degenerate primers have been designed for the detection of these viruses (Briddon and Markham, 1994). With the help of these primers previously uncharacterized geminiviruses can be amplified, and primers designed on the basis of non conserved sequence can be exploited to detect a particular virus and

strain of that virus (McGovern *et al.*, 1994).

(Monga, 2014) reported successful transmission of CLCuV i.e. 91.7per cent in petiole grafting , 83.3 per cent in twig grafting and 100per cent wedge grafting. In wedge grafting technique , the symptoms appeared from 5th day onward and all the tested plants showed symptoms within three weeks, whereas the symptoms appeared between 11-26 and 17-30 days in twig and petiole grafting respectively.

Management of CLCuD

Identification of resistant sources : Though the solution of various diseases is the development of disease tolerant varieties but disease management is quite appropriate when resistance sources are inadequate. In cotton host plant resistance is the best long term and explored strategy to protect the plants from CLCuD (Solomon-Blackburn and Bradshaw, 2007). Singh *et al.,* (2010) reported three genetics stocks namely ABH 47, P 57-6 and F 2164 showed resistant reaction under natural epiphytotic condition but their molecular reaction indicated latent carryover of CLCuD in these symptomless plants. Lines resistant to CLCuD have also reported from Sriganaganagar, HAU, Hisar, PAU, Ludhiana (Anonymous, 2013). But till date due to appearance of recombinant strains of CLCuD, no lines have remained free from the disease.

Through Pollen treatment : Pollen irradiation technique used as a criterion to develop CLCuV tolerant material for creating genetic variability in cotton germplasm. Aslam and Elhai, (2000) used pollen irradiation

Table 1. Losses due to Cotton leaf curl disease (CLCuD) in Cotton under Punjab condition

Year	Cotton Leaf Curl Disease (CLCuD)		Estimated/ expected losses (%)	Remarks
	No. of Location	Per cent disease index visited (%)		
2002	125	7.1	4.80	-
2003	140	30.4	24.9	-
2004	67	3.4	4.17	-
2005	140	2.3	2.13	-
2006	147	5.2	3.57	-
2007	73	8.1	7.21	-
2008	130	1.6	4.01	-
2009	102	43.8	34.8	Very high incidence leaf curl on RCH 134 BGII was observed in Abohar, Fazilka and Kohiwan sarbar block of district Ferozepur.
2010	92	13.5	8.67	-
2011	158	0.80	1.90	Disease incidence was lowest due to frequent rain, which was responsible for causing vector mortality.
2012	38	7.1	7.00	-
2013	31	5.8	4.17	-

technique. They attempted different crosses by applying irradiation doses *i.e.,* 5-10Gy (Aslam and Stelly, 1994) to create more genetic variability.

Cultural and whitefly control : Cotton leaf curl disease spread from the primary inoculum that is present in off season in the form of weeds and other hosts (Monga *et al.*, 2001). The management of CLCuD includes control of vector whitefly and eradication of weeds that contribute the hospitality of Cotton leaf curl virus (Monga *et al.*, 2001). The seed treatment with systemic insecticides may prevent the cotton crop up to 50-60 days. By using insecticides even if infection occurs at later stage the severity of losses may be avoided as symptoms appearance will begin after 65-90 days and plants avoid the most susceptible stage (Singh *et al.,* 2002; Monga *et al.,* 2011). Various agronomic practices like sowing time and application of nutrients (Nitrogen and Potassium) can serve the purpose. The knowledge about K nutrition on association between plants and pests may help in developing strategies to set up high yielding production system by reducing disease incidence (Zafar and Athar, 2013). Choosing best sowing time for a particular variety in different regions is difficult as too early and too late sowing may result in problems of diseases and pests. Appropriate sowing time preferably mid April to mid May results in decrease of disease incidence (Ghazanfar *et al.*, 2007) as compared to delay in sowing from mid May to June. Iqbal and Khan (2010) reported that increased plant spacing in case of early sowing and decreased plant spacing under late sown conditions is effective in management of CLCuD. They also concluded that CLCuD infestation reached its maximum after 105 days of sowing and in case of late sown crop *i.e.* 15 June or later infestation become severe after 45 days of sowing. They recommended 15 cm plant spacing in order to manage CLCuD in case of planting later than 15th of June.

According to Zafar *et al.,* 2010 by understanding the physiological basis of nutrition (nitrogen) strategies can be designed to prevent, escape, avoid and control viral diseases. In case of resistant cultivars nitrogen concentration does not affect but in susceptible cultivars its concentration plays an important role to tackle disease severity. The most recommended management practices to tackle CLCuD disease include virus resistant cultivars, management of causal agents and mineral nutrition (Akhtar *et. al.*, 2004). The influence of Potassium (K) application on disease through specific metabolic functions alters the relationship of host-parasite environment (Kafkafi *et al.*, 2001). Pervez *et al.* (2007) conducted an experiment on role of Potassium (K) in the control of CLCuD. According to their studies by increased application of Potassium up to 250kg/ha results in the reduction of disease from 12 to 38per cent. This increased application contributed considerably as seed cotton yield increased up to 37per cent as compared to Zero-K.

Management through SAR compound : Systemic acquired resistance (SAR) is an induced defence mechanism that contfers long –lasting protection against a broad spectrum of pathogens by the induction of various types of proteins and metabolities (Durrant and Dong, 2004). Studying the protein changes leading to resistance/ tolerance through the application of an elicitor Jasmonic acid (JA) could prove beneficial in disease management (Ryals *et al.,* 1996; Wang *et al.,* 2005). Raj *et al.,* (2014) studied the effect of different doses of Jasmonic acid against CLCud They reported that JA @ 150

uM resulted in lower CLCuD incidence as well as disease index as compared to control in cotton accessions namely RS 921, LH 2076, PIL 8 and Ankur 3028 BGII. Latent carryover detection of symptomless plants treated with 150 uM of JA through PCR amplifications using DNA beta specific primer confirmed the presence of virus in all the tested cotton accession, which showed that induced proteins did not eliminate virus but might be playing role in suppressing the proliferation of virus. Thus JA application resulted in imparting tolerance with induction of protein but did not lead to complete resistance against disease.

Management through biotechnological tools : In plants lacking natural disease resistance PDR approach has been documented to combat different viruses. According to Hashmi *et al.,* (2011) by exploiting transcriptional control two truncated forms of replicase (tACI) gene, capable of expressing only N-terminal 669bp (5'ACI) and C-Terminal 783bp(3'ACI) nucleotides were introduced into *Gossypium hirsutum* through cloning. A strain LBA 4404 of *Agrobacterium tumefaciens* was used through interference technology to impair cotton leaf curl virus in transgenic cotton. When transformed plants were compared with control non transformed plants the over expression of either of the above mentioned nucleotides confer resistance by inhibition of viral genomic and â satellites DNA components. In early and late growth stages Northern blot hybridization revealed high transgene expression (Hashmi *et al.*, 2011).

Future thrusts : Various management strategies of controlling CLCuD can be implemented depending upon conditions. The main emphasis will be concentrated on development of disease resistant varieties/ hybrids based on marker assisted selection (MASS) and trangenics. Mapping the gene (s) conditioning resistance to CLCuD will facilitate their precise and efficient transfer in elite cultivars/ advance lines of American cotton marker assisted selection. Direct crosses of susceptible American cotton lines with desi cotton G. arboretum will be developed employing embryo rescue / ovary culture. Chromosome number of resulting triploids wil be doubled to generate fertile amphidiploids and will be used transfer CLCuD resistance into susceptible American cotton lines. The disease will also be managed by agronomic fertilizer, SAR compound insecticidal control of vector and biotechnological techniques. These methods will be used alone and in combination to manage this severe disease which is still a challenge even after twenty years of extensive research.

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Disease scenario of cotton and management strategies in India- Recent developments

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Historical perspectives and changing disease scenario : The cotton disease scenario has shown a continuous change during the past sixty seven years since independence. When mainly indigenous diploid cottons were being grown in fifties, *Fusarium* wilt, root rot, seedling blight, anthracnose and grey mildew were the major problems. With the large scale cultivation of tetraploid upland cotton (*Gossypium hirsutum*), bacterial blight became the major disease to which indigenous cottons were highly resistant. After the introduction of *Bt* cotton hybrids during 2002 onwards and continuous increase in area under these hybrids to more than 90 per cent of total cotton area till date, the disease scenario has also shown some change. The grey mildew, once a serious problem for diploid cottons especially in central India has now become a major problem in *Bt* cotton hybrids. Grey mildew (percent disease intensity) in central zone was recorded on *Bt* cotton hybrids during 2014-15 in Maharashtra in Akola of Vidahrva region (5.6 to 18.2 % and Nanded-3.9 to 30.5%). In south zone it was severe in two states *i.e.* Karnataka (5-35%), and Andhra Pardesh (0-60.0) during the season. Among other important diseases on *Bt* hybrids, Bacterial blight was reported as important disease in central zone in Maharashtra (Vidahrva- 6.5 to 15.4 %; Nanded 3.5 to 29.7 %) and in south zone in Karnataka (5.0-30.0 %) and Andhra Pardesh (0.0-27.5%). *Alternaria* blight was observed serious during 2014-15 season in Gujrat's Saurashtra area (7.0-22.0%) and Maharashtra's Rahuri (4.4-31.6%) and Nanded (2.0-25.1.%) and in south zone states ie Karnataka (15.0-40.0%), Andhra Pardesh (0.0-53.3%) and Tamil Nadu from 0.0 to 11.0%. (Anonymous, 2015).

Fusarium wilt has become less important as upland cotton now occupying more than 90 per cent area is immune to Indian race of the pathogen. *Verticillium* wilt which appeared in Tamil Nadu remained restricted mainly to that state only.

In north India, the leaf curl disease caused by gemini virus and transmitted by white fly *Bemisia tabaci* has become a threat to cotton cultivation due to development of new recombinant strains and introduction of a number of susceptible *Bt* cotton hybrids in north zone.Severe incidence of disease was observed during 2014-15 season in Punjab(Faridkot-35.0-88.0,Bhatinda-12.7-56,Fazilka-17-80 and Muktsar-10-50), Haryana (Hisar-4.5-76.4,Sirsa-0-26.7,Fatehabad-1.6-24.8, Jind-18.8-41.9 and Bhiwani-0-50.1) and Sriganganagar (7.2-56.6). A disease identified as Tabacco Streak Virus (Ilar virus) transmitted by thrips was observed in the transgenic cotton growing region of Southern Maharashtra and Andhra Pardesh. (Sharma *et al.*, 2007). Avoidable losses due to important diseases like cotton leaf curl virus, (53.6%), bacterial leaf blight(20.6%), Alternaria leaf spot (26.6%), grey mildew(29.2%)and Myrothecium leaf spot (29.1%) have been documented (Monga *et. al.,* 2013) Newer chemicals like propiconazole,

captan+hexaconazole, tetraconazole and strobilurin compounds (fungicides) and copper hydroxide (bactericides) have been successfully tested for the management of foliar disease of cotton (Monga *et. al.,* 2011) Strategies for the integrated management of diseases causing losses in terms of yield and quality need to be redefined.

Role of bioagents and SAR chemicals in disease management : A field experiment was conducted at Dharwad, Guntur and Coimbatore, consecutively for three years during kharif seasons of 2009 to 2012 to evaluate efficacy of the SAR inducing chemicals *viz.,* Salicylic acid, Isonicotinic acid and *Pseudomonas fluorescens* (Pf) strains against foliar diseases of cotton. Eight treatments, comprising of seed treatment and foliar spray with two strains of *P. fluorescens* (CICR H1a and TNAU) and foliar spray of – Propiconazole, – Carbendazim, – Copper oxy chloride plus Streptocycline, Salicylic acid, Isonicotinic acid and – water spray (Control) were evaluated in randomized block design.

Pooled data revealed that the strains of *P fluorescens* applied as seed treatment and foliar spray and propiconazole spray were significantly superior to other treatments against *Alternaria* leaf spot disease at Guntur and Coimbatore. Both *Pseudomonas* strains along with COC + Streptocycline were statistically on par in managing bacterial blight at Guntur. Carbendazim was best in controlling grey mildew at Dharwad as well as Guntur while *Pseudomonas* strains were also superior at Guntur. Propiconazole (0.1%) and COC + Streptocycline were best followed by SAR chemicals against rust at both the centres. Maximum seed cotton yield of 1806 kg/ha was recorded with COC (0.3%) + Streptocycline (0.01%) followed by carbendazim (1765kg/ha) and isonicotinic acid (1763kg/ha). Based on Benefit Cost ratios, it was concluded that the SAR chemicals and Pf isolates can be a good substitute of fungicides for the management of foliar diseases in cotton crop (Bhattiprolu *et al.,* 2014).

Integrated disease management modules : A new programme under AICCIP was initiated during 2011-2012 where studies on developing integrated disease management modules on important diseases prevailing in a particular state were initiated. While designing the experiments, due emphasis was given on bio based interventions and also the new chemicals screened under AICCIP in addition to existing recommended package and practices prevailing in the states. Initially the experiments were designed for Andhra Pradesh, Tamilnadu and Maharashtra (Marathwada region) and three years studies were completed in 2013-2014. Subsequently new experiments were started in Gujrat, Karnataka and Maharashtra (Vidharva region) during 2014-2015 (Anonymous, 2012, 2013 , 2014 and 2015).

Integrated Management studies of cotton diseases (Alternaria Leaf Blight, Bacterial Blight &Rust) at Guntur, Andhra Pardesh for three years (2011 -2013) showed that maximum IBCR of 1.35 was recorded with ST- *Pseudomonas fluorescens*@ 10g/kg seed, SA– *Trichoderma viride* @ 2.5kg/ha, Foliar spray with kresoxim methyl @ 1ml/l at 60 DAS and captan+hexaconazole (Taqat) 1g/l at 90 DAS for fungal diseases in *Bt* hybrid Jadoo BG II.

Integrated management of cotton diseases (Root rot and Alternaria Leaf Blight) at Coimbatore, Tamilnadu (2012 and 2013) showed that the incremental cost benefit ratio with ST – *Bacillus subtilis* (BSC5-TNAU1) + SA @ 2.5 Kg/ha + Foliar spray with *B. subtilis* (1%) on 60, 90 and 120 DAS using Bunny *Bt* was 1.16 as against farmers practice. Likewise the ICBR with respect to the

same module in RCH II Bt was 0.94 in comparison with that of farmers practice.

Integrated Management of *Alternaria* leaf blight (ALB) at Rahuri, Maharashtra (2012 &2013) showed that the seed treatments of bioagent with chemical sprays was found most effective in minimizing the ALB disease intensity by 63.20% with Seed Treatment - PF TNAU1 @ 10g/kg of seed, SA- *T. viride* @ 2.5 kg/ha (TV- TNAU1) in 250 kg of Compost or FYM., Foliar spray with Ergon @ 1ml/L @ 60 DAS and Taqat @ 1.5g/L @ 90 and 120 DAS for fungal diseases in *Bt* hybrid Krishi dhan rakhi.

Based on the studies conducted during 2014-15, at Akola under IDM modules, the seed treatments/soil application of bioagents along with chemical sprays (Seed Treatment – PF CICR @10 g/kg of seed; Soil Application of *Trichoderma viride* @ 2.5 kg /ha TV-TNAU1 and foliar spray with Propiconazol (0.1%) for foliar diseases) were found more effective in minimizing the ALB disease intensity by 67.7 per cent in Bunny *Bt* hybrid. Seed Treatment *Pseudomonas fluorescens* (PF-CICR) @ 10g/kg of seed, soil application *Pseudomonas fluorescens* (PF-CICR) @ 2.5 kg/ha in 250 kg of Compost or FYM and foliar spray with *P. fluorescens* (1%) (PF-CICR) was the best module at Dharwad for the management of seedling mortality and other foliar diseases. Whereas at Junagarh IDM module of seed treatment with *Pseudomonas fluorescens* (PF-CICR) @ 10g/kg seed; soil application with *T.viride* (TV-TNAU) @2.5kg/ha in 250kg of FYM and foliar spray with Ergon @ 1ml/l followed by Taqat @1.5g/l for fungal diseases or COC (0.3%)+ Streptocycline (0.01%) for BLB was best for the reduction of diseases (Alternaria and bacterial blight) and with maximum seed cotton yield.

These studies have indicated that IDM modules using bio based applications in the form of seed treatment/soil application followed by fungicidal interventions can be very fruitful and we therefore need to ensure large scale quality production of such bioagents with the involvement of public/private sector.

New management strategies for cotton leaf curl virus disease : 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* in 1912 by Faquharson. The disease was first reported in India on *G barbadense* at Indian Agricultural Research Institute, New Delhi in 1989. Subsequently it appeared in patches during 1993 around Sriganganagar district of Rajasthan and Ferozepur district of Punjab adjoining to Pakistan border on *G. hirsutum* and spread to entire north Indian cotton zone of around 14 lakh hectares in a short span of 4-5 years. CLCuD is caused by a complex of whitefly (*Bemisia tabaci*) transmitted Begomoviruses having monopartite genome with circular ss DNA associated with satellite (beta and alpha satellite) DNA molecules. Development of resistant/tolerant cultivars, avoidance/eradication of alternate hosts including weeds and whitefly management are the only three effective means of its control.

An experiment with nine interventions *i.e.* buttermilk (the liquid left over after extracting butter from churned yogurt) @ 5 per cent, Cow urine (*Desi* cow) @ 6.6 per cent, *Neem* oil (Azadirachtin-1500ppm) @1 per cent, Mustard oil @ 3 per cent, Kaolin @ 2 per cent, Calcium nitrate @ 0.5 per cent, Potassium nitrate @ 0.5 per cent Paraffin-liquid @ 2 per cent - Kresoxim methyl @ 0.1 per cent, Acephate (chemical control for whitefly) @ 0.4 per cent and Control was conducted for two years (2011-2013) to study their effect on the cotton leaf curl virus disease (CLCuD) and whitefly management. Significantly lower cotton leaf curl virus disease

incidence and percent disease intensity (PDI) compared to control was noted in all interventions except acephate and potassium nitrate. The lowest incidence, however, was observed in cow urine treatment followed by kresoxim methyl, calcium nitrate, buttermilk and neem oil. In case of PDI, it was also the lowest in cow urine treatment followed by kresoxim methyl, *neem* oil, calcium nitrate and buttermilk. The cow urine treatment showed significantly superior and highest seed cotton yield as compared to check. The reduction (%) of whitefly in case of neem oil was 31.19 and 17.52 during 2011-2012 and 2012-2013 whereas Calcium nitrate resulted in 10.49 and 12.84 per cent reduction in whitefly population during 2011-2012 and 2012-2013, respectively. Acephate treated plots showed a reduction of 36.26 and 21.69 per cent during two years of testing. Other treatments did not show any clear and consistent trend. Improvement in Uniformity ratio and fiber strength was noted with various interventions whereas fiber length and micronair were not affected significantly. Validation of results of selected interventions was also carried out in poly house and large plots at experimental area of Regional Station and farmer fields (Monga *et al.,* 2015).

A module with ten treatments and control was tested at four locations(Sriganganagar, Faridkot, Hisar and Sirsa) during 2012 and 2013 to study its efficacy in whitefly and CLCuD management. It was observed that module with sprays of Nimbicidine (30DAS), Admire (45DAS), Nimbicidine (60 DAS), *V. Lacani* (75 DAS), Acephate (90DAS) showed maximum reduction of whitefly. The maximum CLCuD reduction (PDI) was observed when Nimbicidine (30DAS), Cofidore (45DAS), Nimbicidine (60 DAS), V. Lacani (75 DAS) were applied followed by the above referred module which showed maximum whitefly reduction.

A new strategy of screening of these hybrids against CLCuD was evolved under AICCIP. Accordingly, four trials (two normal sown released and pre released *Bt* cotton hybrids with 100 entries each and two late sown released and pre released hybrids with 50 entries each) were planted during 2014-15 with standardized screening protocols using susceptible checks at each location. Based on these trials, CLCuD tolerant hybrids were identified. Similarly, three trials with a total of 130 *Bt* cotton hybrids are laid out at five locations (Hisar, Sirsa, Bhatinda, Faridkot and Sriganganagar) for their screening against CLCuD during the current season ie 2015-16.

Future challenges : Cotton leaf curl virus disease, an important problem presently restricted to north zone need to be dealt more seriously in the context of its possibility of shifting to central and south zone. Further due to changed scenario leading to the development of recombinants and breakdown of resistance, new strategies need to be planned. As no sources of absolute resistance are available in the entire germplasm, we need to look beyond that by introgression of resistance from other available sources through inter specific hybridization. The work on development of transgenics using RNAi technology and marker assisted selection is in progress and may go a long way in development of resistance against this important viral disease. Other components of integrated disease management strategy like cultural practices including weed management and vector control using innovative methods need to be pursued vigorously to obtain a holistic approach. Certain other diseases like *Alternaria*, Bacterial blight and grey mildew showing significant appearance in few areas at present shall need development

of integrated management modules. A vigil is required on the emerging problems like tobacco streak virus and developing protocols of screening methodologies for identification of resistant sources against them. Another important aspect will be to focus on the disease development and progress vis-a-vis climate changes to understand disease epidemiology and fine tune management strategies.

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Integrated disease management of Alternaria leaf spot in cotton

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Cotton is a soft, fluffy staple fiber that grows in a boll or protective case, around the seeds of cotton plants of the genus *Gossypium* in the family of *Malvaceae*. The plant is a shrub native to tropical and subtropical regions around the world, including the America, Africa, and India. The greatest diversity of wild cotton species is found in Mexico, followed by Australia and Africa. Cotton was independently domesticated in the Old and New Worlds.

Current estimates for world production are about 25 million tonnes or 110 million bales annually, accounting for 2.5 per cent of the world's arable land. China is the world's largest producer of cotton, but most of this is used domestically. The United States has been the largest exporter for many years. There has also been a manifold improvement in production, productivity and quality with virtual increase in area. India now produces around 402 lakh bales of cotton ranging from short staple to extra long staple from an area of 122.50 lakh ha with productivity of 537 kg/ha (Anonymous, 2015).

Cotton crop suffers from number of diseases of air, vector and soil borne nature caused by fungal, bacterial and viral pathogens. Geographical origin, distribution and spread of disease is influenced by factors like host, pathogen, environment and external boundaries of the region. The cotton diseases scenario has shown a continuous change during the past sixty five years since independence. Indigenous diploid cottons were mainly grown in fifties and Fusarium wilt, root rot, seedling blight, anthracnose and grey mildew were the major problems. With the large scale cultivation of tetraploid upland cotton (*Gossypium hirsutum*), bacterial blight became the major disease to which indigenous cottons were highly resistant (Monga *et al.*, 1996). After the introduction of *Bt* cotton hybrids during 2002 onwards and continuous increase in area under these hybrids to around 90 per cent of total cotton area till date, the disease scenario has also undergone some change. For instance, the grey mildew, once a serious problem for diploid cottons especially in central India has now become a major problem in *Bt* cotton hybrids in central and south zone in India (Hosagoudar *et al.*, 2008).

However, the production potential of the crop has been not fully exploited due to several biotic and abiotic factors. The crop suffers from many fungal diseases, of which foliar diseases take a heavy toll. Among foliar diseases grey mildew, Ramularia areola, Alternaria blight, *Alternaria macrospora* and bacterial blight *Xanthomonas axonopodis* pv. *malvacearum* are the important ones (Hosagoudar, *et al.*, 2008). Along with this anthracnose caused by *Colletotrichum capsici* and cotton leaf curl virus also cause damage to cotton crop. Cotton leaf rust caused by *Phakospora gossypii* and tobacco streak virus belonging to Ilar group are emerging problems of south zone. The early incidence of rust was

noted in Karnataka and Andhra Pradesh (Anonymous, 2010 and 2011) causing significant losses.

Even before the cultivation of Bt cotton, Alternaria leaf spot of cotton was one of the most important diseases noticed throughout the world. Yield losses upto 26 per cent due to Alternaria leaf spot have been reported (Chattannavar *et al.,*2006).

Plant diseases are considered an important biotic constraint, which leads to significant crop losses worldwide. Integrated disease management (IDM), which combines biological, cultural, physical and chemical control strategies in a holistic way rather than using a single component strategy proved to be more effective and sustainable. In practice and in the majority of cropping systems today, emphasis is still being placed on a single technology. Nevertheless, the use of IDM strategy is gaining momentum, but in developing countries it often lacks the enabling environment for its successful implementation. Success requires appropriate policies in place that cover a wide range of themes such as plant protection, private sector investment, trade and export, food safety, land use, education and awareness, and agriculture extension. Wide adoption of IDM practices is a pre-requisite for achieving impact at the country level. Experience over the last few decades clearly showed that adoption and support for using participatory approaches help farmers improve their overall field management, including disease management, reducing costs and improving production efficiency. In this view several experiments have been carried out for the management of foliar diseases in cotton which have shed light on all the elements that require attention to achieve successful IDM adoption at the national level in developing countries.

COMPONENTS OF DISEASE CONTROL:

Cultural practices : For the control of Alternaria blight, cultural practices such as deep summer ploughing, crop rotation with non host and use of disease free seed, recommended dose of potassium application can help the crop to escape from the disease incidence.

Host plant resistance : Host plant resistance is an important tool to control diseases. The use of resistant varieties is very much welcomed by resource poor farmers because it does not require additional cost and it is environment friendly.

Management of diseases is a continuous process due to development of resistant races of pathogens imposed by climatic changes, chemicals or even resistance to old resistant cultivars. It proved beyond doubt that host plant resistance was most appropriate method compared to any other method as it was the cheapest, safest and most eco friendly approach.

Identification of resistant genotypes is again an essential continuous process either to recommend for cultivation in endemic area or to use as donors of the resistant genes. In view of these, the present investigation of evaluation of cotton hybrids / varieties / genotypes against foliar diseases was carried out to identify the sources of resistance under field conditions by 'Infector Row Technique'. Observations on the intensity of Alternaria leaf spot on each of cotton genotypes / varieties / hybrids were recorded at 90 and 120 days after sowing (DAS). A group of leaves from lower, middle and upper parts from five plants in each genotype was selected randomly and graded according to 0 to 4 scale (Sheo Raj, 1988).

Among 141 cotton genotypes including different *Gossypium spp* Like *G. arboreum* (38), *G. herbaceum* (20), *G. hirsutum* (34), *G. barbadense* (10), *intra hirsutum* hybrids (22) and inter specific hybrid (HxB) (17), evaluated for diseases none of the *G. arboreum* genotypes showed immune and highly resistant reaction to Alternaria leaf spot; however, three genotypes showed moderately resistant reaction and remaining genotypes moderately susceptible to highly susceptible reaction to Alternaria leaf spot disease. None of the *G. herbaceum* genotypes showed immune, highly and moderately resistant reaction to Alternaria leaf spot; however seven genotypes showed moderately susceptible reaction and remaining genotypes showed highly susceptible reaction to Alternaria leaf spot disease. None of the *G. hirsutum* genotypes showed immune, highly and moderately resistant reaction to Alternaria leaf spot; however, eight genotypes showed moderately susceptible reaction to Alternaria leaf spot disease. None of the *G. barbadense* genotypes showed immune, highly and moderately resistant reaction Alternaria leaf spot; however, three genotypes showed moderately susceptible reaction and remaining genotypes showed highly susceptible reaction to Alternaria leaf spot disease. None of the intra-hirsutum hybrid genotypes showed immune, highly and moderately resistant reaction to Alternaria leaf spot; however, eight genotypes showed moderately susceptible reaction and remaining genotypes showed highly susceptible reaction to Alternaria leaf spot disease. None of the interspecific (HXB) hybrid genotypes showed immune, highly and moderately resistant reaction to Alternaria leaf spot; however, three genotypes showed moderately susceptible reaction and remaining genotypes showed highly susceptible reaction to Alternaria leaf spot disease (Table 1).

Numerical rating	Leaf area covered (%)	Reaction
0	Completely free	Immune
2	0-10	Highly resistant
3	11-20	Moderately resistant
4	21-40	Moderately susceptible
5	Ã40	Highly susceptible

Evaluation of *Bt* and conventional cotton entries against foliar diseases of cotton : Among 326 cotton entries (*Bt* and convention entries) evaluated for foliar diseases, none of the entries showed immune reaction to Alternaria blight, 8 entries showed highly resistant, 75 moderately resistant and rest of the lines were susceptible or highly susceptible to Alternaria blight.

Biological control : Use of naturally occurring bio-control agents (antagonists) of plant pathogens can be traced back to many centuries through the traditional practice of crop rotations that primarily permit the reduction of pathogens' inoculum potential in the soil below injury level.

Field experiment was conducted during *kharif,* 2006-2007 at Agricultural Research Station, Dharwad Farm under rainfed conditions for biocontrol. A randomized block design (RBD) with 5 treatments replicated 4 times with a plot size of 5.4 x 4.5 m was adopted. The Abhadita variety was sown during third week of June with a spacing of 90 x 30 cm. Four treatment with bioagents were: Seed treatment with *Pseudomonas fluorescens* formulation (10 g/kg seed) + foliar spray (0.2%) on 30, 40, 50, 60, 70, 80 and 90 DAS, seed treatment with *P. fluorescens* formulation (10 g/kg seeds + foliar spray (0.2%) on 30, 50, 70 and 90 DAS, seed

Table 1. Grouping of cotton genotypes based on their reaction to *Alternaria* blight (*Alternaria macrospora*) under field conditions.

Reaction Rating		Genotypes					
		G. arboreum (38)	*G. herbaceum* (20)	*G. hirsutum* (34)	*G. barbadense* (10)	Intra-hirsutum hybrid (22)	Interspecific Hybrid (HXB)(17)
0	Immune	None	None	None	None	None	None
1	Highly resistant	None	None	None	None	None	None
2	Moderately resistant	CCA4, FDK 173, FDK172	None	None	None	None	None
3	Moderately susceptible	HD466, JT-151, RAAS 36, ARBa 08-34 LD 948, RG 524 PA 255(Quality check) LC, NDLA 2510, T KA 8801, AKA 0109, LD 952, PA 541, 2C (HD123/ AKA 7) / DLSa 17), Ghav 106, ARBa 08- 79 ARBa 08-53 PA 646, CAN 1005, JT 156, CAN 1006, DAS 802, Ghav 109	ANGh 9798/08-2 GShv 630/04, RBDV 31 ANGh 9798/08-3 GBhv 270, ZC (G Cot 23/DDhc 11) GBhv 277	HAG 08-823, NH 633, TSH 9904 SCS 404, RB611, CNDTS 54, NH 632, ARB 08-822	Suvin (CC), DB 1, RHCB 001	CINHH 128, 231, DHH 861, SHH 486, JKCH 2503, MRC 7361, ARCHH 901, DHH 862	GSHB 817, RAHB 302, MCHB 7933
4	Highly susceptible	CISA 104, GAM 158, CINA 357, CISA 3 R , NDLa 2448, RG 542, GAM 138, AKA 0209, ARBa 08-49, CAD 3, CCA 8, JLA 802	DDhc 52, RAHS 31, DDhc 51, GBhv 271, ANGh 9798/08-1 GShv609/04, Bhv 274, RAHS 32, GShv 358/05, GBhv 253	RAH 332, BS 79, LC, NDLH1905, NDLH 1938, SCS 415, CCH 2623, P8608, CPD 812 , L804, AKH016, LH 66, H 1353, ARB 08-4715, GBHV 158, CPD 811, HAG 1602, ZC(LRA/ Sahana), AKH 004, ADB 110, CCH 2629, RB 613, RAH 330, CNH 1104, KH 140 BS 299	CCB 5, DB 11, GSB 41, TCB 108, TCB 47, CCB 1, CCB 6 Bunnyi,	RAHH 331, ARBHH 2062, CAHH 218, BHH 207, SHH 485, LC, GSHH 2342, RAHH 307, Tulasi 144, CINHH 129, ZC (Anlur 651/ ARBHH 2021, BHH 229, NHH 22 5	CCHB 2230, ARBHB 1062, JKCHB 217 DHB 872, Prince, LC, RAHB 301, DCH 32(CC), COCHB 08-1, HAGHB 1064, ARBHB 1040, HAGHB 1036, ARBHB 1021, DCH 871

Table 2. Reaction of *Bt* and conventional cotton entries against Alternaria blight.

Reaction	Genotypes
Immune	None
Highly resistant	KCH 135 *Bt*, DBTHH 01, LD 8, DDCC 1, DLSa 1, DLSa 17, LD 105, LD 102, DT 109,
Moderately resistant	DHH 2078, 2031, 2041, 2050, Bunny, 2036, 2055, 2007, RAH 221, RAHH 95, LD 4, HHB 2, Sahana, MBIPS 105, 104, 206(105), (210) 114, MBIPS 105, SBYF 425, Suvina, Pima, BCS 23, B 82 1 1, IPS 4, IPS 1, IPS 7, Arb 510, Arb 516, 2028, Arb.Chamatkar, 2035 11, Arb 311, Arb 520 6, DHH 11, ACH 155 *Bt*, ACH 21*Bt*, ACH 33 BG II, JK Durga *Bt*, KJ Ishwar *Bt*, MRCH 7201 BG II, PRCH 103 *Bt*, OLE *Bt*, VCH III *Bt*, GK 207 *Bt*, ABCH 1120 *Bt*, Tulsi 4*Bt*, Tulsi 9 *Bt*, Tulsi 117 *Bt*, Dhruv *Bt*, RCH 2*Bt*, RCH 20*Bt*, RCH 111*Bt*, Sandeep *Bt*, Ratna *Bt*, JAI *Bt*, AKKA *Bt*, PCH 930*Bt*, PCH 205 *Bt*, PCH 115*Bt*, NATHBABA *Bt*, Doyor Marnk *Bt*, SP 504 *Bt*, PCH 2270 *Bt*, Rudra *Bt*, NHH 44 *Bt*, Bunny *Bt*, NCS 950, 990, 929, 954, NCHB 992.

Table 3. The effect of *Pseudomonas fluorescens* on seed treatment and foliar spray on Alternaria blight of cotton

Tr. No.	Treatments	PDI		**Mean**	PDC	Yield (q/ha)
		90 DAS	120 DAS			
T_1	Seed treatment with *P. fluorescens* (Pfl) (10 g/kg) + foliar spray (0.2%) on 30, 40, 50, 60, 70, 80 and 90 DAS	24.31 (29.52)	18.33 (25.34)	**21.32**	34.00	1975.09
T_2	Seed treatment with *P. fluorescens* (Pfl) (10 g/kg) + foliar spray (0.2%) on 30, 50, 70, and 90 DAS	25.63 (30.41)	21.24 (27.14)	**23.44**	27.43	1883.32
T_3	Seed treatment with *P. fluorescens* (Pfl) (10 g/kg) + foliar spray (0.2%) on 30, 60, and 90 DAS	29.21 (23.80)	24.21 (29.46)	**26.71**	17.30	1633.73
T_4	Copper oxychloride (0.3%) + Streptocycline sulphate (0.05%)	33.39 (30.34)	27.21 (31.42)	**30.30**	6.19	1666.66
T_5	Control	35.79 (34.23)	28.80 (32.32)	**32.30**	-	1029.11
	SE m+	0.318	0.471	-	-	134.41
	CD (p=0.05)	0.982	1.412	-	-	400.70

* Figures in parentheses indicate angular transformed values

PDI : Per cent disease index PDC : Per cent disease over control AB : *Alternaria* blight DAS : Days after sowing

treatment with *Pseudomonas fluorescens* formulation (10 g/kg seeds + foliar spray (0.2%) on 30, 60 and 90 DAS, and Copper-oxychloride (0.3%)) + Streptocycline Sulphate (0.05%)). Untreated plot was maintained as control. Disease grading was done using 0-4 scale (Sheo Raj,1988). Percent disease incidence was calculated using Wheeler's formula (1969) and finally per cent disease control was calculated. Yield was recorded in each treatment separately.

Seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 40, 50, 60, 70, 80 and 90 DAS recorded significantly low Alternaria blight per cent disease index (21.32 PDI) followed by seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%)

on 30, 50, 70 and 90 DAS (23.44 PDI), seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 60 and 90 DAS (26.71 PDI) and Copper-oxychloride 0.3%) + Streptocycline Sulphate 0.05%) (30.30 PDI) were least effective. Seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 40, 50, 60, 70, 80 and 90 DAS showed maximum per cent disease over control of 34.00 PDC followed by seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 50, 70 and 90 DAS (27.43 PDC), seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 60 and 90 DAS (17.30 PDC) and Copper oxy-chloride (0.3%)) + Streptocycline Sulphate (0.05%)) (6.19 PDC).

The kapas yield obtained from different treatments was significant. The maximum yield (1975.09 kg/ha) was recorded in seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 40, 50, 60, 70, 80 and 90 DAS treatment followed by seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 50, 70 and 90 DAS (1883.32 kg/ha) treatment followed by Copper-oxychloride 0.3%) + Streptocycline Sulphate 0.05%) (1666.66 kg/ha) and seed treatment with *P. fluorescens* (10 g per kg seeds + foliar spray (0.2%) on 30, 60 and

Details of the treatments under investigation

Tr. No.	Treatments
T	*T. harzianum* (TH-KSD), seed treatment (0.8%) + foliar spray (0.8%) at 65, 85 and 105 DAS
T^1	*T. viride* (TV 97), seed treatment (0.8%) + foliar spray (0.8%) at 65, 85 and 105 DAS
T^2	*T. harzianum* (SK1), seed treatment (0.8%) + foliar spray (0.8%) at 65, 85 and 105 DAS
T^3	*P. fluorescens* (A), seed treatment (0.8%) + foliar spray (0.8%) at 65, 85 and 105 DAS
T^4	*Azadirachta indica,* seed treatment (10%) + foliar spray (10%) at 65, 85 and 105 DAS
T^5	NSKE, seed treatment (10%) + foliar spray (10%) at 65, 85 and 105 DAS
T^6	Nimbicidine, seed treatment (0.4%) + foliar spray (0.4%) at 65, 85 and 105 DAS
T^7	Propiconazole foliar spray (0.1%)
T^8	Copper oxychloride foliar spray (0.3%)
T^9	Control
T_{10}	

Table 4. Field efficacy of bio agents and botanicals on incidence of Alternaria leaf spot, yield and yield parameters of *Bt* cotton (*kharif* 2010-2011)

Tr.No.	Treatments	PDI	PDC	Average bolls/ plant	Yield (kg/ha)
T_1	*T. harzianum* (TH-KSD), ST (0.8%) + FS (0.8%)	11.51	67.70	26.47	2451.13
T_2	*T. viride* (TV-97), ST (0.8%) + FS (0.8%)	10.30	71.10	27.20	2485.84
T_3	*T. harzianum* (SK1), ST (0.8%) + FS (0.8%)	11.13	68.77	27.07	2466.05
T_4	*P. fluorescens* (A_1), ST (0.8%) + FS (0.8%)	13.0	63.30	26.00	2393.72
T_5	*Azadirachta indica*, ST (10 %) + FS (10%)	20.12	43.55	24.47	2173.15
T_6	NSKE, ST (10 %) + FS (10%)	17.06	52.13	25.60	2206.17
T_7	Nimbicidine, ST (0.4 %) + FS 0.4%)	12.78	64.14	26.20	2400.00
T_8	Propiconazole, FS (0.1%)	8.33	76.63	28.07	2670.37
T_9	Copper oxychloride, FS (0.3%)	9.37	73.71	27.87	2500.90
T_{10}	Control	35.64	-	23.20	2012.41
	CD (p=0.05)	2.66		2.83	253.52

ST – Seed treatment , FS – Foliar spray

90 DAS (1633.73 kg/ha, Table 3).

Management of Alternaria blight using bio agents and botanicals : The investigations were under taken to study integrated management of Alternaria leaf spot disease on *Bt* cotton through bio-agents and botanicals. For this purpose field experiments were conducted during *kharif* seasons of 2010-2011 and 2011-2012 at Agricultural Research Station, Dharwad Farm under rainfed conditions, to find out efficacy of bio agents and botanicals against Alternaria leaf spot of *Bt* cotton under field conditions. Per cent Disease Index (PDI) and per cent disease control (PDC) were determined 120 days after sowing following Wheeler (1969).The yield and fibre quality were also recorded and economic analysis was worked out. The data obtained were statistically analysed following the procedure given by Panse and Sukhatme (1985).

The results obtained during 2010-2011 (Table 4) revealed that Propiconazole foliar spray (FS) (0.1%)) significantly lowered Alternaria leaf spot per cent disease index (PDI) to 8.33 per cent which was at par with *T. viride* (TV-97) seed treatment (ST) (0.8%)) + foliar spray (FS) (0.8%)) with 10.30 %) PDI followed by *T. harzianum* (SK1), ST (0.8%)) + FS (0.8%)) with 11.13 PDI. *Azadirachta indica* leaf extract ST (10%) + FS 10%) was the least effective with 20.12 %) PDI. Maximum per cent disease control (PDC) among bio agents and botanicals was recorded with *T. viride* (TV-97), ST (0.8%)) + FS (0.8%)) with 71.10 %) PDC, followed by *T. harzianum* (SK1), ST (0.8%)) + FS (0.8%)) with 68.77 %) PDC, and *T. harzianum* (TH-KSD), ST (0.8%)) + FS (0.8%)) with 67.70 %) PDC respectively. Significantly maximum average bolls/plant were recorded with Propiconazole, FS (0.1%)), which was *at par* with bio agents and botanicals.

The fiber (*kapas*) yield among the treatments significantly varied. However, the maximum yield of 2670.37 kg/ha was recorded in Propiconazole, which was *at par* with *T. viride* (TV-97), ST + FS (2485.84 kg/ha), *T. harzianum* (SK1), ST + FS (2466.05 kg/ha) and *T. harzianum* (TH-KSD), ST + FS (2451.13 kg/ha). The fiber quality was statistically *at par* among all treatments. Almost similar results were obtained during 2011 – 2012 (Table 5).

The pooled data (Table 6) revealed that Propiconazole foliar spray significantly lowered Alternaria leaf spot PDI, which was *at par* with *T. viride* (TV 97) followed by *T. harzianum* (SK1 and TH-KSD), while *Azadirachta indica* leaf extract ST + FS was least effective. The nine treatments of bio agents, botanicals and fungicides tested significantly reduced the least per cent disease index of Alternaria leaf spot disease over control. Significantly maximum pooled average bolls/plant were recorded in case Propiconazole, which *at par* with *T. viride, T. harzianum,* and *P.fluorescens* while significantly least average number of bolls/plant were recorded in case *Azadirachta indica* leaf extracts.

The variation in pooled cotton fiber (kapas) among the treatments was significant. However, the maximum yield of 2721.27 kg/ha was recorded with Propiconazole, *T. viride* (TV 97), (2543.73 kg/ha) and *T. harzianum* (2528.10 kg/ha) (Table 5). Chattannavar *et al.* (2001) observed that the treatment with *P. fluorescens* gave 39.72 per cent disease control of Alternaria blight, while *P. fluorescence* gave 37.26 per cent disease control. Chidambaram *et al.* (2004) tested *T. viride* Miller and *T. harzianum* Rifai and two strains of bacterial biocontrol agent *P. fluorescens viz.,* Pf1 and CHAO in field trials for their bio efficacy against Alternaria leaf spot disease of cotton. They observed that all bio-control agents were significantly effective. The effectiveness of biocontrol agents was comparable to that of chemical fungicides like Propiconazole 25 EC and

Table 5. Field efficacy of bio agents and botanicals on incidence of *Alternaria* leaf spot, yield and yield parameters of *Bt* cotton (*kharif* 2011-2012)

Tr.No.	Treatments	PDI	PDC	Average bolls/ plant	Yield (kg/ha)
T_1	*T. harzianum* (TH-KSD), ST (0.8%) + FS (0.8%)	10.10	69.81	28.33	2552.51
T_2	*T. viride* (TV-97), ST (0.8%) + FS (0.8%)	8.99	73.13	29.20	2601.62
T_3	*T. harzianum* (SK1), ST (0.8%) + FS (0.8%)	9.58	71.37	28.67	2590.15
T_4	*P. fluorescens* (A_1), ST (0.8%) + FS (0.8%)	13.32	60.19	28.47	2501.10
T_5	*Azadirachta indica*, ST (10 %) + FS (10%)	18.74	43.99	26.13	2248.26
T_6	NSKE, ST (10 %) + FS (10%)	16.45	50.84	26.73	2365.66
T_7	Nimbicidine, ST (0.4 %) + FS (0.4%)	11.64	65.21	27.93	2501.74
T_8	Propiconazole, FS (0.1%)	7.14	78.66	31.33	2772.18
T_9	Copper oxychloride, FS (0.3%)	8.83	73.61	29.60	2621.97
T_{10}	Control	33.46	-	24.47	1919.07
	CD (p=0.05)	2.51		2.92	260.28

ST – Seed treatment FS – Foliar spray

Table 6. Field efficacy of bio agents and botanicals on incidence of *Alternaria* leaf spot and yield parameters of *Bt* cotton (pooled data of *kharif* 2010-2011 and 2011-2012)

Tr.No.	Treatments	PDI	PDC	Average bolls/ plant	Yield (kg/ha)
T_1	*T. harzianum* (TH-KSD), ST (0.8%) + FS (0.8%)	10.81	68.73	27.40	2501.82
T_2	*T. viride* (TV-97), ST (0.8%) + FS (0.8%)	9.65	72.08	28.20	2543.73
T_3	*T. harzianum* (SK1), ST (0.8%) + FS (0.8%)	10.36	70.03	27.87	2528.10
T_4	*P. fluorescens* (A_1), ST (0.8%) + FS (0.8%)	13.20	61.79	27.23	2447.41
T_5	*Azadirachta indica*, ST (10 %) + FS (10%)	19.43	43.76	25.30	2210.71
T_6	NSKE, ST (10 %) + FS (10%)	16.76	51.51	26.17	2285.92
T_7	Nimbicidine, ST (0.4 %) + FS (0.4%)	12.21	64.66	27.07	2450.87
T_8	Propiconazole, FS (0.1%)	7.74	77.61	29.70	2721.27
T_9	Copper oxychloride, FS (0.3%)	9.10	73.66	28.73	2561.43
T_{10}	Control	34.55	-	23.83	1965.74
	CD (p=0.05)	1.95		1.98	202.49

ST – Seed treatment FS – Foliar spray

Copper oxychloride 50 WP.

Chemical control: For many decades fungicides played an important role in disease control. In the 1960s, systemic fungicides started gradually to replace the older non-systemic chemicals with more effectiveness and specificity in disease control. The availability of a variety of new products, with narrow and broad specificity, offer important disease control options, however, their practical application continues to face the risk of selection of resistant pathogen populations. Experience accumulated over the last few decades clearly showed that fungicidal application had a better impact when used within an IDM strategy.

A field experiment was conducted during *kharif* 2006-07 at Agricultural Research Station, Dharwad Farm under rainfed conditions. A randomized block design (RBD) with four treatments replicated five times with a plot size of 5.4 x 4.5 m was adopted. The Abhadita variety was sown during third week of June with a

spacing of 90 x 30 cm. Two fungicides i.e., Propineb (0.1%) and 0.2%)) and Propiconazole (0.1%)) were sprayed thrice at an interval of 15 days starting from the initial appearance of the disease. Untreated plot was maintained as control. Disease grading was done using 0-4 scale (Sheo Raj, 1988). Percent disease incidence was calculated using Wheeler's formula (1969) and finally per cent disease control was calculated. Yield was recorded in each treatment separately.

Propineb (0.2%) recorded significantly low Alternaria blight per cent disease index (20.61 PDI) followed by propineb 0.1 per cent (24.92 PDI) while propiconazole 0.1 per cent (25.99 PDI) was least effective. The three treatments of fungicides tested significantly reduced the maximum per cent disease over control in case of propineb 0.2 per cent (23.27 PDC) followed by propineb 0.1%) (7.22 PDC) and propiconazole 0.1 per cent (3.23 PDC). These results are in conformity with the reports of Chattannavar *et al.* (2000a, 2006).

The kapas yield variation among the treatments was non-significant. However, the maximum yield of 1752.99 kg per ha was recorded in propineb 0.2 per cent followed by propineb 0.1 per cent (1735.46 kg/ha) and propiconazole 0.1 per cent (1680.65 kg/ha) (Table 7).

Avoidable losses due to cotton diseases in India : Studies carried out under All India Coordinated Cotton Improvement Project over the years at selected locations have demonstrated avoidable losses due to important diseases like cotton leaf curl virus, bacterial leaf blight, Alternaria leaf spot, grey mildew, *Myrothecium* leaf spot, *Helminthosporium* and rust. Among them, Alternaria leaf spots can cause loss upto 26.6 per cent based on results (2006- 2007 to 2008-2009) of study conducted in central India at Rahuri and south zone locations at Guntur and Dharwad. The result indicated that five sprays of Propiconazole (0.1%) at 35, 50, 65, 80, and 95 DAS decreased percent disease index (PDI) from 31.59 to 20.85 per cent thereby reducing yield loss due to Alternaria leaf spots in variety LRA-5166. The implication of these studies in disease management is given (Table 8).

A field experiment was conducted at Agricultural Research Station, Dharwad farm, Dharwad, Karnataka during *Kharif* 2009-2010 and 2010-2011 to evaluate the field bio-efficacy of triazole group of fungicides against major fungal foliar diseases grey mildew caused by *Ramularia areola* Atk. and Alternaria blight or Alternaria leaf spot caused by Alternaria

Table 7. The effect of fungicide foliar sprays on the severity of Alternaria blight in cotton

Tr. No.	Treatments	PDI		**Mean**	PDC	Yield
		90 DAS	120 DAS			(kg/ha)
T_1	Propineb (0.2%)	22.10 (28.02)	19.11 (25.88)	**20.61**	23.27	1752.99
T_2	Propineb (0.1%)	28.62 (32.34)	21.23 (27.39)	**24.92**	7.22	1735.46
T_3	Propiconazole (0.1%)	25.84 (30.53)	26.14 (30.72)	**25.99**	3.23	1680.65
T_4	Control	26.00 (30.64)	27.72 (31.79)	**26.86**	-	1628.06
	SE m+	0.226	0.946	-	-	66.162
	CD (p=0.05)	0.696	2.838	-	-	NS

* Figures in parentheses indicate angular transformed values

PDI: Per cent disease index PDC: Per cent disease over control AB: *Alternaria* blight DAS: Days after sowing

Table 8. Crop loss due to *Alternaria* leaf blight (Rahuri, Guntur and Dharwad; Pooled 2007-2009)

Treatments	PDI	Yield(q/ha)	Yield loss
Propiconazole(0.1%) at 35DAS	28.43	13.30	14.17
Propiconazole(0.1%) at 35, 50DAS	27.34	13.12	15.35
Propiconazole(0.1%) at 35, 50 and 65DAS	26.14	14.13	8.79
Propiconazole(0.1%) at 35, 50, 65 and 80DAS	24.41	14.05	9.37
Propiconazole(0.1%) at 35, 50, 65, 80 and 95DAS	20.85	15.50	0.00
Propiconazole(0.1%) at 50, 65,80 and 95DAS	20.56	14.56	6.06
Propiconazole(0.1%) at 65, 80 and 95 DAS	22.08	13.36	13.80
Propiconazole(0.1%) at 80 and 95 DAS	24.01	12.81	17.37
Propiconazole(0.1%) at 95 DAS	26.88	12.94	16.47
Control	31.89	11.38	26.59

macrospora Zimm were compared with standard recommendation Carbendazim 50%) WP foliar spray treatment (0.1%). The experiment was planned in Randomised block Design and replicated thrice on Bt cotton hybrid "Bunny Bt". The individual treatment plot size was 6.0 x 5.4 m^2 with spacing of 90 x 60 cms. Normal recommended cultural practices were adopted. Three sprays of all treatments were undertaken immediately after the appearance of the disease at an interval of 12 days. The observations on percent disease index of Alternaria blight and grey mildew were recorded 15 days after the last spray, on five randomly selected plants in each treatment. In each treatment, ten plants were randomly selected and tagged. Three branches were randomly tagged per plant and the intensity of Alternaria blight and grey mildew on all the leaves of these tagged branches were graded by adopting 0 to 4 scale as given by Sheo Raj (1988).

Efficacy of triazoles in management of major foliar diseases of cotton : The results obtained during 2009 with respect to Alternaria blight, revealed that, all the treatments were significantly superior over untreated control. From the data, it is clear that, the treatments *viz.,* Penconazole, Difenconazole and Hexaconazole were found on par with each other with PDI of 5.80, 6.70 and 8.30, respectively and they were significantly superior to all other treatments followed by Propiconazole, Mycobutanil, Tridemefon with PDI of 9.40, 10.30 and 11.40 respectively. The results obtained during *kharif*, 2010 followed similar trend of results but in slightly higher intensity of incidence of disease, as observed during *kharif*, 2009 (Ashtaputre *et al.*, 2011).

The pooled data (Table 9) of two years for Alternaria blight, indicated that all the treatments were significantly superior over untreated control. The triazoles under study were found to be significantly effective in the management of the diseases. The least PDI was observed in Penconazole of 6.10 PDI for Alternaria blight followed by Difenconazole (7.10 PDI for *A.* blight) and Hexaconazole (8.20 for A. blight) which were *on par* with each other and significantly superior over rest of the treatments followed by all other triazole group of fungicides under study.

Cotton yield : The cotton yield was significantly superior in all the treatments as compared to untreated control. The results indicated that, all the triazoles under study have showed higher yield. Next best treatments were *viz.,* Tridemorph (13.9 q/ha), Propineb (13.6 q/

ha), Carbendazim (13.5 q/ha) and Mycobutanil (13.2 q/ha), *on par* with each other, but differed significantly with the untreated control.

The pooled data of two years depicted that, the triazole group of fungicides was found to be more effective in enhancing the yields significantly (Table 9). Pooled maximum yield of both the years was noticed in Penconazole (16.4 q/ha), which was significantly superior over all other treatments, followed by Difenconazole (15.2 q/ha), Propiconazole (14.9 q/ha), Triadimefon (14.7 q/ha) and Hexaconazole (14.5 q/ha).The least yield was noticed in untreated control (11.9 q/ha). All the treatments were found to be significantly differ with untreated control.

Benefit cost ratio (BCR) : From the pooled data of two years, it is evident that maximum B:C ratio was observed in Hexaconazole (9.63) followed by Propiconazole (6.2) and Penconazole (5.80) (Table 9).

In the present investigation, it is evident that all triazoles under study were found to be effective in control of Alternaria blight disease, which in turn reflected in more cotton yield. Among these triazoles, Penconazole followed by Hexaconazole and Difenconazole reduced the disease severity of the disease effectively and also enhanced the yield. These findings are in accordance with Khodke and Raut, 2009, who reported that these triazoles gave the effective control of grey mildew.

The benefit cost ratio is an important parameter for recommendation of any treatment for successful control of plant disease. In the present study, though the treatments containing three sprays of Penconazole, Hexaconazole, Difenconazole, Triadimefon and Propiconazole gave significant control of both the diseases, maximum Cost Benefit ratio of 9.63 was realized in treatments containing three sprays of Hexaconazole (0.1%)) followed by Propiconazole (6.2) and Penconazole (5.80). This clearly indicated that three sprays of Hexaconazole (0.1%)) are more useful not only in reducing the

Table 9. Efficacy of triazoles against Alternaria blight of cotton

	Treatments	PDI		**Pooled mean PDI**	Yield (q/ha)		**Pooled mean yield (q/ha)**	B:C
		2009-2010	2010-2011		2009-2010	2010-2011		
T1	Mycobutanil @ 1gm/litre	10.30(18.73)*	11.20(19.53)	**10.70(19.13)**	12.80	13.7	**13.2**	1.81
T2	Hexaconazole @ 1ml/ litre	8.30(16.67)	8.20(16.59)	**8.20(16.63)**	14.30	14.8	**14.5**	9.63
T3	Penconazole @ 1ml/ litre	5.80(13.93)	6.40(14.60)	**6.10(14.27)**	17.20	15.6	**16.4**	5.80
T4	Propiconazole @1ml/ litre	9.40(17.90)	9.30(17.73)	**9.40(17.82)**	14.10	15.7	**14.9**	6.2
T5	Difenconazole @ 1ml/ litre	6.70(15.03)	7.40(15.77)	**7.10(15.40)**	14.80	15.5	**15.2**	3.27
T6	Tridimefon@ 1gm/ litre	11.40(19.70)	11.20(19.57)	**11.30(19.63)**	14.10	15.3	**14.7**	3.12
T7	Tridemorph@ 1ml/litre	19.90(26.50)	20.00(26.58)	**20.00(26.54)**	13.60	14.2	**13.9**	4.83
T8	Carbendazim @1gm/litre	20.10(25.67)	21.30(27.50)	**20.00(26.58)**	13.20	13.9	**13.5**	5.77
T9	Propineb @3gm/litre	18.20(25.27)	17.00(24.35)	**17.60(24.81)**	12.90	14.2	**13.6**	3.65
T10	Control	30.30(33.37)	33.00(35.08)	**31.60(34.22)**	11.20	12.7	**11.9**	-
	SEm±	1.187	1.068	**0.748**	0.351	0.389	**0.219**	
	CD (p=0.05)	3.527	3.174	**2.222**	1.041	1.157	**0.651**	

*Figures in parentheses are arcsine values

cost of protection but also gave higher benefits as compared to other treatments and can be recommended as one of the components in integrated disease management of cotton. This is followed by Difenconazole and Penconazole applications. Similar types of findings are observed by many workers (Khodke and Raut, 2009, Algarsamy and Tagarajan,1986). Hence, spraying of Hexaconazole (0.1%) could be considered as an effective management practice to manage major fungal foliar diseases.

CONCLUSION

The ultimate aim of promoting IDM is to empower its users to engage in competitive and sustainable agricultural production with long-term positive impacts on poverty and human and environmental health. The impact of successful implementation of IDM approaches beyond pilot scale cannot be achieved if IDM is not strategically positioned within national policies for agricultural production and protection, and within the broader context of agricultural and rural development, human and environmental health.

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Impact of nematode disease on the cotton production in India

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Ever since the dawn of civilization, cotton has played a major role in weaving social, economic and political fabric of our country. Still India holds the distinction as the world's second largest producer and consumer of cotton after China. A large number of rural families in 10 states depend on cotton for their livelihood. Unfortunately, despite having maximum area under cotton cultivation, productivity of cotton in India at 552 kg lint / ha lags (*www.cotcorp.gov.in/statistics.aspx*) far behind as against productivity of 2281 kg lint / ha in Australia *(www.indexmundi.com › Agriculture).* By the year 2030 based on current demand and projected growth demand for cotton is expected to be 48 MTons (Kranthi *et al.,* 2011). The demand for cotton is likely to increase in future with the enhanced consumer awareness and preference for natural fibers compared to manmade ones . The projected demand for cotton fiber can be met by two pronged approach - by increasing per ha productivity of cotton and ameliorating losses caused due to biotic factors. The strategy is to optimize environment in which crop is grown and neutralize damaging biotic factors so that inherent yield potential comes close to realization.

Loss estimates : Plant parasitic nematodes, are the hidden enemy of crops with the estimated overall annual yield loss of world's major crops due to damage by phytoparasitic nematodes reported to the extent of 12.3 per cent (Sasser and Freckman, 1987). Nematode problems are exacerbated in the tropics as climate conditions are ideal for nematode development and are now compounded by agricultural practices as monoculture of susceptible cultivars that favor population development and thus crop damage. The national loss due to plant parasitic nematodes in 24 different crops in monetary terms has been worked out to the tune of 21068.73 million rupees (Jain *et al.*, 2007). About 19 Genera of plant parasitic nematodes have been recorded for their association with cotton. In Indian context three nematodes Root-knot nematode *Meloidogyne incognita*, reniform nematode *Rotylenchulus reniformis,* Lesion nematode *Pratylenchus spp.* and lance nematode *Hoplolaimus columbus* have been recognised as most important.

Root knot nematode *Meloidogyne incognita* : The root knot nematode, *Meloidogyne incognita* is the most pathogenic species with a host range spanning over 300 plant genera in India. A recent survey in the districts of Haryana and Punjab has revealed widespread infestation of root knot nematode in *Bt* cotton crop in north India**.** The root knot nematode fusarium wilt disease complex is becoming prevalent now even in *hirsutum* cotton**.** Yield losses of 16 to 25 per cent caused by *M. incognita* on cotton have been reported in Haryana (AICCIP, 2012). On National scale cotton crop losses ranging between 12.3-20.8 per cent have been attributed to *M. incognita* (Khan *et al.*, 2010a). Of six races of *M.incognita*

documented so far (Robertson *et al.,* 2009), only race three and four are known to attack cotton. Race diversity of *M. incognita* across India has been recorded and race two, three and five have been reported predominantly on different crops in Maharashtra (Darekar and Mhase 1988, Khan, 1997, Khan *et al.,* 2014). Race three is reported from Karnataka and Tamilnadu on cotton (Krishnappa,1985) while race four has been recorded on cotton from north India (Verma and Jain,1999). Though in Maharashtra reniform nematode is predominant on cotton (Gokte-Narkhedkar,1999), severe infestation of root knot nematode, *M. incognita* on several vegetable crops as well as cotton growing in vicinity was encountered in Nagpur, Wardha, Yavatmal and Chandrapur regions (Pers.). Based on differential host test and molecular charactrizaton, these were found to belong to race 3. Phylogenetic Tree using Neighbour Joining method for multigenes of ribosomal RNA showed that Nagpur population is distinct from other populations from Maharashtra.

Reniform nematode, *Rotylenchulus reniformis* : Reniform nematode (*R. reniformis*), first described from Hawaii, USA is widespread in the tropics and subtropics. The Reniform nematode has a wide host range spanning 115 plant species in 4 families. There also exist two races *i.e.* A and B for Reniform nematode. However, only race' A' attacks cotton. Nematode causes delay in boll maturity, as well as the reduction in boll size and lint quantity. It has long been suspected that there exist more races of Reniform nematode than thought of earlier, however confirmation is yet to be done. It is also reported to cause an increase in Wilt disease development in wilt susceptible varieties. *R. reniformis* has been noticed as the most frequent and dominant species in cotton growing areas of central India. Populations of *R. reniformis* showed two peaks, a low during summer and high in autumn. Work done at CICR, Nagpur has put threshold level for reniform nematode on cotton at 1 nematode per cc soil.

Work done at CICR, Nagpur indicated increase in yield by 8 to 10 per cent in nematicide treated plots over the untreated control. Field trials on avoidable yield losses conducted at CICR regional Station, Coimbatore showed yield increase by 9.5 to 17.4 per cent when the nematicide Metham sodium (Vapam, Sistan) was applied. In India, crop loss due to Reniform nematode (*R. reniformis*) on cotton has been put at14.7 per cent. Damages to cotton by *R. reniformis* has been studie in USA and estimated at 5.6 per cent due to direct reduction in yield, lint percentage and reduced fiber elongation.

R. reniformis has been noticed as the most frequent and dominant species in cotton growing areas of central India. Populations of *R. reniformis* showed two peaks, a low during summer and high in autumn. Yield loss due to reniform nematode depends on soil conditions, especially the soil moisture, cultivar tolerance and type of soil. It has been observed that diffusates released by germinating seedlings serve to attract nematodes. The nematode penetration occurs all along the root except the root tip. Optimal soil moisture for reproduction ranges between 25- 30 per cent and *R. reniformis* can survive without host for more than 25 months. Even in air dried soil (3.3 % moisture) survival for 7 months at 20- 30°C is reported. Soil pH is also an important factor affecting reproduction of the nematode and reniform enamtode thrives best in slightly acidic soils with optimum pH between 4.8 and 5.3. Work done at CICR, Nagpur also indicated that phenomenon of phased infectivity occurs in reniform nematode

. Individuals of population pool of reniform nematode exhibited phenomenon of staggered infectivity. This has significance that in case of invading juveniles not completing their life cycle, a small percentage of population always remains to ensure survival and perpetuation

In recent years reniform nematode problem has been reported from certain areas as Buldhana, Akola etc. where cotton is grown as monoculture under drip irrigation and nematode population were at 2-5 nematodes/ cc soil, much above the threshold level of damage. Infected plants showed stunting with yellowing leaves. Root system of affected plants was also affected.

Lance nematode, *Hoplolaimus Columbus :* Though lance nematode *Hoplolaimus* Columbus has been recorded on cotton in many areas of central India, very little information is available on its damage potential. Noe (1993) has recorded that 10 per cent loss in cotton occurs due to initial inoculum level of 70/100 cm^3 soil. Infestations of H. columbus may suppress yields of cotton 10-25 per cent (Mueller and Sullivan, 1988; Noe *et al.*, 1991)

Lesion nematode,*Pratylenchus sp* As the name suggest the most characteristic symptom of lesion nematode *Pratylenchus* spp is the appearance of lesions on the roots which initially look as tiny elongated water soaked spots.The lesions enlarge gradually, coalesce, and ultimately girdle the root giving an appearance of constriction. The lesions are formed due to release of hydrolytic enzymes during feeding. In general, necrotic symptoms are limited to feeding site. Browning of roots also occurs due to the accumulation of phenolic compounds in areas of injury. Infected plants show moderately to severely stunted plants in discrete patches with yellowish to chlorotic leaves. Stunting of crop, general loss of vigour and gradual wilting are also associated in lesion nematode complex. *Pratylenchus thornei* has been reported with cotton in many areas of central India. However their damage potential is still not worked out. (Gokte Narkhedkar *et al.,* 2002)

Diagnosis of nematode problem in cotton : The nematode problems in cotton is suggested by one or more of the following: 1) Cropping history of the field, *e.g.* continuous two or more years production of cotton or equally nematode-susceptible crops; Monoculture of cotton makes strong case for nematode problems particularly in irrigated tracts 2) Above ground symptoms including off colour leaves and stunted cotton in spots or large areas of a field. General foliar symptoms of nematode damage may include areas or spots in the field where plant size is reduced. Height and size of cotton are often reduced, and this is generally seen as irregular growth in oval patterns. Plants also prematurely wilt on hot days and sometimes take on an off green cast. Heavily infested fields, particularly those with reniform nematodes, will show widespread irregular cotton plant growth (wavy appearance). Severely damaged cotton plants often show nutrient deficiency symptoms which can complicate diagnosis

Why nematode infestation on *Bt* cotton : Bacteria *Bacillus thuringeinsis* is known to be antagonistic to plant parasitic nematodes. *Bt* produces crystal proteins that also target nematodes (Wei *et al.*, 2003). *Bt* toxins cause suppression of nematode reproduction, egg hatching and juvenile survival, as well as direct killing of nematodes (Siddiqui and Mahmood, 1999). In laboratory studies, eggs and Juveniles exposed to different *Bt* isolates at 50 per cent

concentration of cell free filtrate recorded inhibited egg hatching of eggs and mortality of second stage juveniles. (Khan *et al.,* 2010b). In other studies, purified *Bt* toxin did not have any toxic effect on RKN and inoculated plants had a higher galling index than the uninoculated plants (Devidas and Rehberger, 1997). There are six proteins (Cry5, Cry6, Cry12, Cry13, Cry14, Cry21) known to be toxic to larvae of a number of free living or parasitic nematodes (Kotze *et al.*, 2005).Cry5B also contains four of five sequence blocks conserved among most Cry toxins suggesting that it folds into 3D structure identical to Bt toxins Cry1Ab. Soil bioassay evidence that Cry1Ab and Cry3Bb1 toxins, at higher than field relevant doses, have an inhibitory effect on the growth and reproduction in *Caenorhabditis elegans* (Hoss *et al.,* 2008, 2011). However, the widespread infestation of rootknot and reniform nematode in *Bt* cotton areas points towards inefficacy of *Bt* toxin expressed by transgenic crops. The susceptibility of transgenic cotton to RKN has also been reported by Colyer *et al.* (2008) .. In other studies, purified Bt toxin did not have any toxic effect on rootknot nematode and on the contrary inoculated plants had a higher galling index than the uninoculated control plants (Devidas and Rehberger, 1997). *Bt* genes (Cry 1Ac, Cry2Ab and their combinations) deployed for bollworms–complex management do not seem to have any effect on plant parasitic nematodes**. (**Lingaraju *et a*l., 2012)**.** Al-Deeb *et al.,* (2003) and Hoss *et al.*, (2011) also concluded that nematode abundance and functional diversity were not significantly affected in rhizosphere soil of MON88017 or MON863 *Bt* corn. A. Recent study (Karuri *et al.,* 2013) reported that Bt cotton expressing Cry1Ac and Cry2Ab2 protein was more susceptible to *M. incognita* than its isoline. also reported that *Bt* cotton containing Cry1Ac and Cry2Ab2 protein had no significant effect on nematode diversity. The question is why no effect on nematode populations despite *Bt* toxins being toxic to nematodes .

Cry proteins or pleiotropic effects resulting from genetic transformation may have been responsible for the susceptibility of Bt cotton to *M. incognita* as alterations in the host plant may change the chemicals responsible for nematode attraction including feeding behavior. Other limiting factor could be the size of toxin proteins. Results have shown that 54 kDa Cry6A protein can be ingested by *M. incognita*. The size exclusion limit for *H. schachtii* was approx. 23 kDa (Urwin *et al.*, 1998), but the upper limit for other nematodes had not been confirmed. Size of Bt toxins cry1Ac, cry2Ab is about 65kda. It is also possible that due to size limit less toxin is ingested by plant parasitic nematodes and transgenic cotton shows nematode infestation..

Amelioration of nematode problems :

Lack of effective nematicides along with concomitant dangers of environmental pollution limits use of chemicals for nematode control. Resistant varieties are not available and crop rotations designed to reduce nematode density are not economically practicable. Soil solarization wherever feasible should be practiced for nematode management. There is also need for exploration of novel methods for management of nematodes and genetic modification mediated crop protection fits the bill.

Researchers in different laboratories are deploying two approaches in use of GM plants for nematode management . Transgenic expression of a range of proteinase inhibitor, most of them occurring naturally in plants, is being tried for nematode control. This approach relies on transgenic plants expressing

proteinase inhibitor which prevents digestion of dietary plant proteins by the feeding nematodes. Specific root specific promoters have been developed to ensure the inhibitor is expressed only in roots where nematodes feed . This strategy prevents root invasion without a direct lethal effect to nematodes and suppresses the nematode population build up. Research work at Leeds, UK has provided proof for effectiveness of this strategy with cysteine protease inhibitors (cystatins) in crops as rice, potato and banana. Expression of protease inhibitors in transgenic crops were recorded to confer resistance to the potato cyst nematodes, G*lobodera pallida* and *G. rostochiensis,* whilst on root knot and other nematodes efficacy was variableTransgenic plants expressing insecticidal toxins, peptide repellants, acetylcholine receptor antagonist among others have also potential for nematode control. Lectin toxicity is attributed to their ability to interfere with intestinal function. They are highly resistant to proteolysis in the gastrointestinal tracts and are therefore available to bind glycans on the surface of the intestine interferring with intestinal function. GNA- Snowdrop lectins are broad spectrum but effect on nematodes vary.

The other approach is post-transcriptional gene silencing (PTGS) of nematode parasitism genes through delivery of double stranded RNA (dsRNA) from the host. Using RNA interference, the nematode's biology can be turned against itself resulting in endogenous nematode control. RNAi mediated gene silencing provides a strategy for developing nematode resistant crops for which natural resistance genes do not exist or are not very effective. Both the approaches need not be exclusive and in combination may offer greater durability and efficacy against nematodes.

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Status of nematode problems on cotton : An Indian scenario

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Cotton is a major fibre crop of global importance and has high commercial value. It is grown in temperate and tropical regions of more than 70 countries and is grown in all types of soil except pure sand, saline and water logged soils. Specific areas of its production include India, China, US, Pakistan, Uzbekistan, Egypt, Turkey, Australia, Greece etc. Cotton, popularly known as **"White Gold"** is an important crop in world agriculture and is used by about 75 per cent of world's population for textile purposes because its fiber is used universally as a textile raw material. In addition to clothing, cotton seed is a major source of vegetable oil and cotton cake is a rich source of high quality protein for stock feed. In India, it is important cash and commercial crop valued for its fibre and vegetable oil and thus earning valuable foreign exchange by providing employment to millions of people, hence plays a significant role in national economy.

India continued to maintain the largest area under cotton and is second largest producer of cotton next to china with 35.3 per cent and 24.0 per cent of world cotton area and production, respectively. India also sustained the position of being the second largest consumer and exporter of cotton. In India, the major cotton growing states are Punjab, Haryana, Maharashtra, Madhya Pradesh, Gujarat, Tamil Nadu, Karnataka etc. and the area under cotton cultivation during 2013-2014 was 11.7 million hectares with production around 29 million bales of 170 kg. As regards cotton production, Gujarat is the leading cotton producing state with 106.8 lakh bales. The state of Haryana had cotton in an area of 5.57 lakh hectares with production of 20 lakh bales of 170 kg during 2013-2014 (All India Coordinated Cotton Improvement Project – Annual Report, 2013-2014).

The cultivation of cotton falls into three main groups *viz.*, Egyptian or American Egyptian type, the long staple cotton, *i.e.*, *Gossypium barbadense* (allotetraploid), American and African upland medium staple cotton, *i.e.*, *G. hirsutum* (allotetraploid) and Asiatic old world short staple cotton, *i.e.*, *G. arboreum* (diploid) and *G. herbaceum* (diploid). *G. hirsutum* accounts for over 80 per cent of global cotton production. But, during past few years, it has been observed that traditional cotton cultivation of non *Bt* has given way to cultivation of *Bt* cotton hybrids.

The successful raising of this crop is hampered by the attack of number of insect pests and diseases. During recent past, phytoparasitic nematodes, which not only cause diseases by themselves directly but also aggravate the disease caused by fungi, have assumed significance in limiting the production of cotton in many cotton growing areas of the country. Because cotton is grown as a cash crop, it is often grown in monoculture system that favours the development of a nematode community that is dominated by one or a few species of plant parasitic nematodes. The important nematode pests of cotton include root-knot nematode (*Meloidogyne incognita*), reniform nematode (*Rotylenchulus reniformis*), lance nematode

(*Hoplolaimus* spp.), sting nematode (*Belonolaimus longicaudatus*) and a few other nematodes of minor importance such as lesion nematode (*Pratylenchus* spp) and stubby root nematode (*Trichodorus christiei*) (Edward *et al.*, 1993). Root-knot nematodes are major problem in many cotton growing areas of India while reniform nematode has regional importance.

Economic importance of plant parasitic nematodes in cotton : The annual yield loss in cotton due to phytoparasitic nematodes on worldwide basis is reported to be 10.7 per cent (Sasser and Freckman, 1987). Among these parasites, the root-knot nematode, *Meloidogyne incognita* is a major yield limiting factor in many countries including India. Davis and May, 2005, recorded the yield suppression in cotton which ranged from 18.0-47.3 per cent in 2002 and from 8.5-35 per cent in 2003 by the southern root-knot nematode.. Similarly, a loss of 17.0 per cent in cotton has been attributed to root-knot nematodes alone in Brazil (Sasser, 1979). Equally important was the reniform nematode. Even as high as 40-60 and 9.5-17.4 % avoidable yield losses were estimated after application of nematicides in Egypt and India, respectively, due to *Rotylenchulus reniformis* (Oteifa, 1970, Palanisamy and Balasubramanian, 1983) while Robinson (2007) estimated annual loss to U.S. cotton crop to be $ 130 million and stated that this nematode has replaced the root-knot nematode as the major nematode of cotton in few US states. The avoidable losses in cotton yield due to root-knot nematode (*Meloidogyne incognita*) under field conditions in Haryana ranged from 16.8 to 20.0 per cent (Jain *et al.*, 2000).

ROOT KNOT NEMATODE (*MELOIDOGYNE* SPP)

A. Distribution : : Among various species of root-knot nematodes, *Meloidogyne acronea and Meloidogyne incognita* are known to parasitize cotton. However, *M.incognita* (Kofoid and White, 1919) Chitwood, 1949, is the only species which is of consequences to the cotton throughout India. It is worldwide in its distribution from nearly all cotton growing areas especially where soils are coarsely textured (Starr et al., 1993). *M. acronea* has so far been found only in some areas of South Africa. The earliest report of root-knot nematode on cotton was from USA (Atkinson, 1989). However, in India the first report of its occurrence on cotton was by Luthra and Vasudeva (1939) from Punjab followed by Thirumalachar (1946). Besides India, it has also been reported from Taiwan (Tu *et al.*, 1972); Pakistan (Tanveer and Haq, 1975); Egypt (Ibrahim *et al.*, 1979); USSR (Khurramov, 1982); Brazil (Lordello *et al.*, 1984); South Africa (Wyk *et al.*, 1987); USA (Martin *et al.*, 1994); China (Yang, *et al.*, 1992), Kenya (Karuri *et al.*, 2010) and from many other countries.

Prasad (1960) observed *M.incognita* on cotton at IARI, New Delhi, Abu Bucker and Seshadri (1968) reported it, attacking cotton from Tamil Nadu, Darekar *et al*. (1992) from Maharashtra, Patel (1984) from Gujarat reported *M.incognita and M.javanica* attacking cotton. Sakhuja *et al.* (1986) reported it from Punjab and race 4 of *M.incognita* infecting cotton in Sirsa district of Haryana was reported first time from India and later on by others (Bajaj *et al.,* 1986; Vats *et al.,*1999, Verma and Jain, 1999). However, presence of race 3 of *M.incognita* equally capable of damaging cotton has been reported from Tamil Nadu and Karnataka (Krishnappa, 1985). The frequency of occurrence of root-knot nematode (*Meloidogyne incognita* race 4) in major cotton growing areas of Haryana was however high (Vats *et al.*, 1999).

B. Symptoms : The foliar symptoms of root-knot nematode attack on cotton are not very diagnostic. Infestation results in uneven, pale, stunted and sick crop. The general symptoms of damage include dwarfing, chlorosis and temporary wilting and a general unthrifty appearance giving the look of nutritional deficiency symptoms. High population density of the nematode at sowing can kill the plants at seedling stage. Cotton is a fairly drought resistant crop by virtue of its long tap root which may reach depths of more than one meter and any damage to this tap root can severely restrict the uptake of water and nutrients, leading to loss of vigour in rest of the plant. Root-knot nematodes attack both tap and lateral roots and leads to formation of galls/knots thereby causing disruption in meristematic zone, which may lead to slowing down or even complete cessation of tap root growth depending upon the initial nematode population in the soil. However, galls on cotton are not as big and numerous as on other susceptible crops like vegetables.

C. Life history/cycle : The life cycle of *M.incognita* is slightly prolonged on cotton when compared to other hosts. The second stage juveniles of root-knot nematode penetrate the roots usually in first 2 cms of root tip and thereafter, they migrate to stelar region. The comparative penetration of *Meloidogyne incognita* in cotton and tomato roots showed 11.3 and 17.2 per cent larval penetration respectively. One generation was completed in 30 days on tomato compared to 32 days on cotton (Rai and Jain, 1989). The life cycle lasts for 33-38 days at 25æ% C on *G. barbadense* and for 32 days on *G. hirsutum* (Rai and Jain, 1989).

The pathogenicity of *M.incognita* on cotton revealed that there was significant decrease in plant growth characters at and above one j_2/g soil in non *Bt* cotton (Rai and Jain, 1989) and *Bt* cotton (Chawla, 2015). Further, the reproduction factor (Rf) of *M.incognita* infecting cotton under different soil textures showed maximum Rf value under sand while clay and clay loam soils were least favoured in American cotton (Verma, 1997) as well as in *Bt* cotton (Chawla, 2015). Histopathological changes in cotton roots due to the feeding of *M.incognita* showed that nematode feed on xylem tissue and giant cells were formed in metaxylem. Nematodes were observed lying parallel to stellar region forming multinucleate giant cells with dense cytoplasm (Rai and Jain,1989).

D. Disease complexes : Besides, inflicting direct damage to the plants, root-knot nematodes interact with other micro-organisms such as fungi, bacteria, viruses etc. and thus form disease complexes as phytophagous nematodes are part of soil microfauna like other organisms. In all these interactions, there is greater incidence of wilt or seedling disease with greater yield suppression when cotton is exposed to multiple pathogens than when only a single pathogen is present. Interaction of *M.incognita* with root-rot fungi *Rhizoctonia solani* revealed that concomitant occurrence of both the pathogens led to higher damage to cotton plants as compared to their individual effect. However, maximum damage was recorded when *M.incognita* was inoculated one week prior to *R.solani* in cotton (Verma, 1997). However, when fungus was inoculated one week before nematode, the nematode penetration was restricted upto cortical region. Carter, 1975 concluded that root-knot nematodes were debilitating parasites which weakened the plants and made it more susceptible to attack by *Rhizoctonia* spp. The posterior end of the nematode body was seen out of the epidermis and fungus presence was seen

in the form of sclerotia in the cortical tissue (Tekchand *et al.*, 1992). The severity and incidence of wilt of cotton due to *Fusarium oxysporum* f.sp. *vasinfectum* has also been found to increase in presence of *M.incognita*.

E. Nematode management:

I. Chemical methods: Basically, two types of chemicals *viz.,* fumigants and non-fumigants have been used. Soil fumigation with ethylene dibromide, dichloropropene-dichloropropane (DD) and dibromochloropropane (DBCP) proved effective but, DBCP was most promising. However, due to their prohibitive cost, difficulty in application methods, environmental and toxic hazards, their use has been banned in most of the countries including India. Thereafter, non fumigant (non volatile) chemicals were begun to be used for controlling phytonematodes infecting cotton. These chemicals mainly belong to carbamates (aldicarb and carbofuran) and organophosphates (phorate, phenamiphos etc.) group. In Punjab, application of carbofuran @ 1 kg a.i./ha at sowing followed by additional dose of 2 kg a.i./ha 50 days thereafter led to 41 per cent higher cotton yield in root knot nematode affected fields (Sakhuja *et al.,* 1987). In order to use the chemical pesticides, judiciously, seed treatment with systemic nematicides has been found to be an effective proposition, which gives protection for 3-4 weeks thereby providing healthy start to the plant. Seed soaking treatment with monocrotophos or carbosulfan each @ 2000 ppm for two hours or use of neem based pesticides *viz.,* Achook, Nimbicidine etc. @ 1, 2 and 4 per cent have shown promising results against *M.incognita* infecting cotton (Vats *et al.,*1997 and 1998) or a novel nematicide, abamectin @ 100g/ 100 kg seed (Monfort *et al.,* 2006). It has been observed that carbamates like aldicarb and carbofuran gave better results than organophosphatic compounds like phorate and disulfoton in controlling nematodes attacking cotton (Kumar and Agarwal, 1985).

II. Cultural methods : Harnessing of solar energy in northern part of India, where maximum temperature many a times, go as high as 48°C, can help in controlling these noxious soil borne pathogens. Deep summer ploughing of nematode infested fields not only leads to disturbance and instability in nematode community but also causes mortality by exposing the nematodes to solar heat and desiccation and hence reduction in their initial population.

Azadirachta indica leaves used 20 g/kg soil proved effective in terms of minimum galling (16.7 per plant) compared to 118.3 per plant in untreated check. Further, of the various organic manures *viz.,* neem cake, poultry manure, spent compost, FYM and biogas slurry, proved best in improving growth parameters of cotton (Vats *et al.*, 1998, Verma and Jain, 2001). The crop rotations with wheat, barley and oat and also corn and soybean have shown promising results in containing the buildup of root-knot nematode population (Singh *et al.*, 1998, Koenning and Edmisten, 2008).

III. Biological control: Soil application or seed treatment with the number of micro-organisms have been found effective to suppress root-knot nematode infestations in cotton. Various bio agents like oviparasitic fungi, *Paecilomyces lilacinus* and rhizospheric bacteria have been investigated for controlling *M.incognita* infecting cotton. *Azotobacter chroococcum* used as seed dressing treatment method in cotton in *M.incognita* infested soil (Lakshminaryanan *et al.*, 1995) and *Pseudomonas fluorescens* (Timper

et al., 2009) mitigated the adverse effects of nematode infection by reducing galling and egg mass production. *Gluconacetobacter diazotrophicus* st. 35-47 (rhizobacteria) used as seed treatment, accounted for 35-47 per cent higher cotton yield in *M.incognita* infested fields (Bansal *et al.,* 2005). This practice has been included as a recommendation for adoption by the farmers in the Package of Practices of *kharif* crops in Haryana. Further, use of vesicular arbuscular mycorrhiza (VAM) fungi (*Glomus fasciculatum*) when applied in *M.incognita* infested soil was effective in increasing plant growth and reducing nematode galling and multiplication (Verma and Jain, 2004). Even a complex of *M. incognita* and root rot fungus, *R. bataticola* was managed successfully using *Trichoderma viride* (Verma, 2011) while Verma and Nandal, 2009 observed reduction in root knot nematode population on cotton using *T. viride* and 50 kg P/ha.

IV. Host plant resistance : Quite a good number of genotypes have been screened by various workers for resistance against *M.incognita*. But, so far no promising genotypes, which could be used for transferring resistance into commercially cultivated type is yet available. However, the cotton variety Auburn 623 has been found to possess a high level of tolerance to root-knot nematode races. Fa LSS and Arkot 9111 have exhibited resistance against *M.incognita* (Dube *et al.*, 1988, Bourland and Jones, 2005). As far as *Bt* cotton is concerned, high degree of susceptibility to root-knot nematode has been reported in all hybrids screened (Verma *et al.,* 2011, Chawla, 2015).

V. Integrated management : For the management of this nematode, Verma *et al.,* (2011) recorded highest increase in *Bt* cotton yield (39.5%) over check and significantly lowest final nematode population in treatment combination of *Gluconacetobacter diazotrophicus* strain 35-47 @ 50 ml/5kg seed and carbofuran @ 1.0 kg a.i./ha as soil application followed by 29.3 per cent yield increase over check in seed treatment with carbosulfan (3.0%) w/w + soil application of carbofuran @ 1.0 kg a.i./ha. Seed dressing treatment with carbosulfan (3.0%) (w/w) alone or with soil application of sebufos @ 1.0 kg a.i./ha at sowing proved effective (Vats *et al.*, 2000). Similarly, Chawla, 2015, observed maximum growth of *Bt* cotton plants and minimum number of eggs and final nematode population in soil treatment with either carbofuran or neem cake + seed coating with carbosulfan.

RENIFORM NEMATODE (*ROTYLENCHULUS RENIFORMIS*)

A. Distribution : This nematode was first observed on the roots of cotton in Baton Rouge, Lousiana (Smith and Taylor, 1941). This is the only species of this nematode to cause economic losses in cotton. It is primarily an inhabitant of tropical and subtropical areas including U.S., (Jones *et al.*, 1959), Egypt (Oteifa, 1970) and India (Varaprasad, 1986; Abu Bucker and Seshadri, 1968). Das and Gaur, 2009 have recently reported high frequency of occurrence of this nematode from Punjab (56.5%), U.P. (42.3%) and Haryana (30.0%). Unlike *M. incognita*, *R. reniformis* is favoured by fine textured soils with a relatively high content of silt or clay (Robinson *et al.*, 1987). In addition to all types of cotton, wide spread occurrence of the nematode was reported in Bt cotton hybrids (Banu, 2009).

B. Symptoms : The nematode causes dwarfing, chlorosis, premature decay and loss of

secondary roots and plant mortality. Injury to cotton becomes evident with reduction in emergence of seedlings. The roots of such plants are smaller, fewer and sometimes show browning. Affected plants bear lesser and smaller bolls. There is delay in maturity, a reduction in size of boll and lint percentage, plant growth, seed index and fibre micronaire value (Jones *et al.*, 1959). Stunted patches in fields having pale greenish leaves occur due to the infestation of this nematode in gardenland cotton in Tamil Nadu. Patel *et al.*, 1996 observed an initial inoculum level of 1000 nematodes/plant and above to be pathogenic on cotton.

C. Life history/Cycle: Two races (A and B) of *R. reniformis* have been reported from India, out of which only race A infects cotton (Dasgupta and Seshadri, 1971). The first stage juvenile moults in the egg and comes out as second stage juvenile. The second, third and fourth stage do not feed. After fourth moult, the immature females penetrate the roots and become reniform (Kidney shaped) in five days. The nematode shows no specificity to the age of plants roots but they prefer succulent roots on which they feed close to the root tip. They feed on phloem tissues and cause necrosis of phloem and its parenchyma. Damage to epidermal and parenchyma is observed near site of infection and feeding is limited. Males also penetrate but do not feed. It took 17-23 days for the completion of one life cycle on cotton (Birchfield, 1962). Life table studies of this nematode revealed that the approximate generation time was 25-45 days. Life cycle duration in *Bt* cotton hybrids and their non transgenic parents was found to be similar (Banu, 2009)

D. Disease complexes: The first indication of the association of *R. reniformis* with *Fusarium* wilt of cotton was observed as early as 1940. This nematode forms disease complexes with *F. oxysporum f. sp. vasinfectum*, *Verticillium dahliae* and with several seedling disease pathogens in which this nematode increases the incidence and severity of seedling disease on cotton (Brodie and Cooper, 1964). Studies on the interaction between *R. reniformis* and *R. bataticola* (virulent and avirulent strains) revealed that both strains of the fungi were equally effective in causing seedling rot of cotton in the presence of this nematode (Patel *et al.*, 2004)

E. Nematode management:

I. Chemical methods: Soil application of carbofuran @ 0.5 kg/ha gave good results against reniform namatode in Brazil. Its population was significantly reduced by seed treatment with carbofuran @ 2.0 per cent (w/w) coupled with soil application of either carbofuran, phorate, phenamiphos or carbosulfan each @ 1.0 kg/ha which gave maximum reduction in nematode population at 30 days after germination with significantly higher cotton yield over control (Patel *et al.*, 1996). Soil application of FYM @ 25t/ha together with carbofuran @ 1.0 kg/ha managed reniform nematode effectively and increased seed cotton yield. Robinson *et al.*, 2002, had reported that due to the depth of distribution of this nematode in some soils, yield response to fumigation can be improved by deeper placement of 1,3-dichloropropene.

II. Cultural control: Crop rotation has been reported to be effective in suppressing densities of *R. reniformis* despite the nematode's wide host range. Although crop rotation with non-host crops such as corn and soybean are effective

in reducing population and damage incurred by this nematode, rotation with these crops are often economically prohibitive (Davis *et al.*, 2003). A number of non-host crops are known for this nematode (Varaprasad *et al.,* 1986) but their suitability with reference to the biotype existing in a particular locality and economic considerations remains to be ascertained. Mustard and Sesamum, in cropping sequence, reduce populations in Northern India. *Crotolaria juncea* grown as antagonistic crop also suppressed this nematode (Marla *et al.*, 2008)

III. Biological control: Successful suppression of the nematode on cotton by nematophagous, *Pochonia chlamydosporia* was reported by Wang *et al.,* 2005 under green house conditions. Seed and soil application with *Pseudomonas fluorescens* reduced the nematode population to the tune of 63.5 per cent. Similarly, a complex of this nematode with root rot fungus was managed successfully by seed and soil treatment of *T.viride* and *P. fluorescens* (Sivakumar, 2009).

IV. Host plant resistance: Use of cultivars resistant to reniform nematode would become a major component of nematode management program in cotton. High level of résistance to this nematode has been found in *G. arboreum* Nanking CB 1402, *G. barbadense* Texas 110, *G. somalense* and *G. stocksii* (Carter, 1981).

LANCE NEMATODE (*HOPLOLAIMUS* SPP) : Five species of lance nematode *viz., H. columbus*, *H. galeatus, H. indicus*, *H. seinhorsti* and *H. aegypti* have been reported to be pathogenic to cotton. In India, *H. indicus* and *H. seinhorsti* were reported by Abu Bucker and Seshadri, 1968 and Verma and Jain, 1999. Field studies in US showed that *H. columbus* could reduce cotton yields upto 19.0 per cent (Noe and Imbriani, 1986). Heavily infested cotton manifests severe stunting, yellowing and almost complete defoliation in *H. galeatus* infestation. All species exhibit both endoparasitic and ectoparasitic feeding habits. The nematodes start feeding on young tap roots of cotton immediately after seedling emergence (Brodie and Cooper, 1964). Cavities are formed on the root cortex due to destruction of cells. The nematodes also penetrate epidermis but do not cause extensive damage as they do in cortex. Tyloses are formed in the affected xylem resulting in plugging of the xylem elements (Lewis *et al,* 1976). The life cycle of *H. indicus* from egg to egg is completed at 27-36 days at a temperature of 28 to 32* C. Hussey (1977) reported control of lance nematode in the southern US by aldicarb or DBCP treatment and or sub soiling to a depth of 5 cm which resulted in deeper penetration by cotton roots.

Few other nematodes such as sting nematode (*Belonolaimus longicaudatus*), lesion nematode (*Pratylenchus* spp) and stubby root nematode (*Trichodorus christiei*) are of minor importance and hence are not covered under this chapter.

Futuristic approaches : Besides root-knot nematodes, populations of other nematodes is also recorded from some areas. Hence, there is need to carry out intensive surveys for recording other economically important phytonematodes associated with cotton. During recent times, the cotton cultivation has boosted due to introduction of *Bt* cotton hybrids which replaced conventional non *Bt* cultivars at national and state level, so the research work involving *Bt* cotton becomes necessary as all the previous work was conducted on non *Bt* varieties. Except some preliminary work on *Bt* cotton, the

systematic and in-depth research on *Bt* cotton was necessitated as now the cotton production and profitability has increased by adoption of *Bt* cotton Other points of future thrusts can be:

- Carry out studies on host parasite relationships.
- Carry out studies on nematode interactions with other co-habiting microorganisms.
- Intensive survey on nematodes on *Bt* cotton in India has to be carried out.
- Screening of transgenic plants to nematodes.
- Identification of resistance genes to reniform and root-knot nematode in cotton.
- Conduct studies on INM in cotton.

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Molecular characterization of upland cotton (*Gossypium hirsutum* L.) by using RAPD markers

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Abstract : The present study attempts to evaluate 15 genotypes of G. *hirsutum* L. to study the genetic diversity by using Random Amplified Po lymorphic DNA (RAPD) analysis. Polymerase chain reaction (PCR) was carried out by using 25 random decamer primers. Twenty one primers were found polymorphic and produced 149 bands with 7.0 bands per primer. The polymorphism percentage ranged from 20 to 100 per cent. The genetic similarity coefficient for all genotypes ranged from 0.65 to 0.86 per cent. Cluster analysis separated in to three clusters which were corresponded well with their centers or sub centers or genetic relationship. The first major group comprised three cultivars (KH 120, KH 121 and L 765), cluster II comprised of three sub clusters (II A, II B and II C). Cluster II A comprised three cultivars (DHY 286 IR, MCU 5 and L 761). Cluster II B comprised 4 cultivars (NH 545, NH 572, PH 297 7 1 and PH 348). The cluster II C comprised three varieties (PH 44 1 2, PH 1009, PH 1024). The cluster III comprised single genotype *i.e.* Cocker showed 0.66 similarity index. The present study is also an endeavor in that direction and the information generated from it will useful to utilize in future cotton breeding programmes.

Key words : Cotton, PCR, RAPD

Cotton (*Gossypium hirsutum* L.) is currently the leading plant fibre crop worldwide and is grown commercially in the temperate and tropical regions of more than 50 countries. It is fibre, oil and protein yielding crops plays a crucial role in the economy of India. For multiple use of lint and byproducts cotton is also referred as "White Gold". A classical breeding has contributed tremendously in terms of quality and yield, further improvements in yield, fibre strength, length, water absorption and thermal properties are required for textile and other industrial applications. The potentials for improving these properties through classical breeding are limited. New technological advancement over the last decades in the field of genetics and plant breeding has provided the superior tools for detailed genetic analysis of agricultural crops. These considerations have led to the exploration of other techniques like DNA profiling. Molecular markers allow the breeder to dissect complex traits without exhaustive field screening over time and space, thus avoid the unreliable phenotypic assays. DNA markers are successfully applied in identification of genotypes with better fibre quality genes, which can fetch good price to Indian farmers (Waghmare *et al.*, 2005).

Among several molecular techniques, Randomly Amplified Polymorphic DNA *i.e.* RAPD is widely used due to technical simplicity and comparative high speed of technology. It is useful technique for construction of genetic linkage map because amplified DNA fragments originating from segregating population are inherited as dominant or recessive genes

following the classical Mendelian pattern. RAPDs are dominant markers which discriminate the individuals on the basis of presence or absence of particular RAPD band (Livneh and Vardi, 1998). A present study was conducted in attempt to study genetic diversity within 15 cotton genotypes.

MATERIALS AND METHODS

The genomic DNA of 15 cotton genotypes *viz.,* KH 120, DHY 286 IR, PH 297 7 1, KH 121, NH 572, L 765, L 761, PH 348, PH 330, PH 44 1 2, PH 1009, PH 1024, NH 545, Cocker, MCU 5 were used to study their genetic diversity. The genotypes are grown in plastic tray pots in a polyhouse. DNA was isolated by CTAB (Cetyle Tetra Methyl Ammonium Bromide) method described by Zhang and Stewart, (2000). Polymerase chain reaction (PCR) was carried out by using 25 random decamer primers. The amplified product of RAPD from Agarose gel images were scored for presence (1), absence (0). Data analysis was performed using NTSYS PC (Numerical Taxonomy System, Version 2.02). The SIMQUAL programme was used to calculate the Jaccard's coefficient. Dendrogram was constructed using unweighted pair group method for arithmetic mean (UPGMA) based on Jaccard's coefficient. The polymorphic percentage of the obtained bands were calculated by using formula, Polymorphic % = (Number of polymorphic bands/ Total bands) x 100.

RESULTS AND DISCUSSION

The genomic DNA from 15 cotton genotypes were evaluated to study the genetic diversity by using Random Amplified Polymorphic DNA (RAPD) analysis. Polymerase chain reaction (PCR) was carried out by using 25 random decamer primers. Perusal of data revealed that total 149 bands were observed with 7.0 bands per primer. RAPD analysis which revealed that twenty one primers were found polymorphic and four primers *viz.,* OPK 2, OPK 06, OPA 14 and OPA 19 were failed to amplify.

Banding pattern generated by primers

i) OPK 01, OPK 07, OPA 05 and OPA 20 : Total six bands each were produced by these primers, out of which five were polymorphic and one was monomorphic. Molecular polymorphism produced by these primers was 83.00 per cent.

ii) OPK 03 : Five bands were generated by OPK 03, four bands were polymorphic and one was monomorphic in nature. Level of polymorphism was 80.00 per cent.

iii) OPK 04 : Amplification product of OPK 04 accounts for five bands. One band was polymorphic and four were monomorphic. Thus level of polymorphism was 20.00 per cent.

iv) OPK 05 : OPK 05 primer generated eight bands out of which seven bands were found polymorphic. Thus OPK 05 primer showed 87.50 per cent polymorphism.

vi) OPK 08, OPK 09 and OPA 10 : The primers OPK 08, OPK 09 and OPa 10 generated seven bands each; out of seven bands six bands were polymorphic in nature. Polymorphism percentage was 85.71 per cent shown by these primers.

vii) OPA 06 : Ten bands generated by OPK 06, nine were polymorphic while one band was monomorphic in nature. Thus primer estimated 90.00 per cent polymorphism.

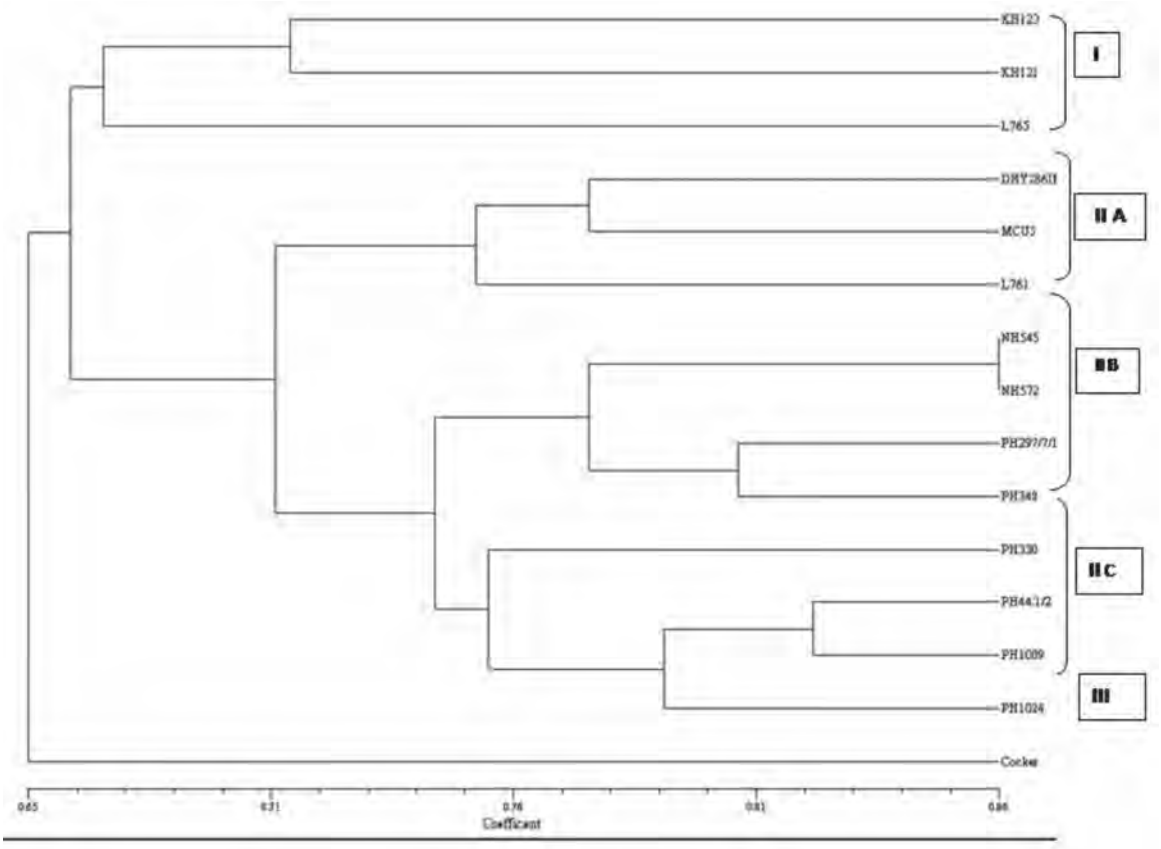

Fig. 1. Dendrogram showing the genetic similarity among 15 cotton genotypes as derived from RAPD data using the UPGMA

vii) OPA 07, OPA 08, OPA 11 and OPA 16 : The primers OPA 07, OPA 08, OPA 11 and OPA 16 had generated nine bands each, all these bands were polymorphic. No any unique or monomorphic band was observed. Molecular polymorphism percentage was accounted to 100 per cent.

viii) OPA 09 and OPA 17 : Total six bands each were generated by OPA 09 and OPA 17 three were polymorphic and three were monomorphic, thus produced 50 per cent polymorphism.

ix) OPA 12 : Total seven bands were produced by OPA 12, out of which four were polymorphic, three were monomorphic. Molecular polymorphism produced was 57.14 per cent.

x) OPA 13 : Total eight bands were produced by OPA 13, out of which three were polymorphic, five were monomorphic. The percentage of polymorphism was 37.50 per cent estimated by the primer OPA 13.

xi) OPA 15 and OPA 18 : Nine bands each were generated by the primer OPA 15 and OPA 18, six were polymorphic in nature. However three monomorphic band was observed. The percentage was 66.67 per cent projected by these primers.

It is evident from Table 2 that twenty one

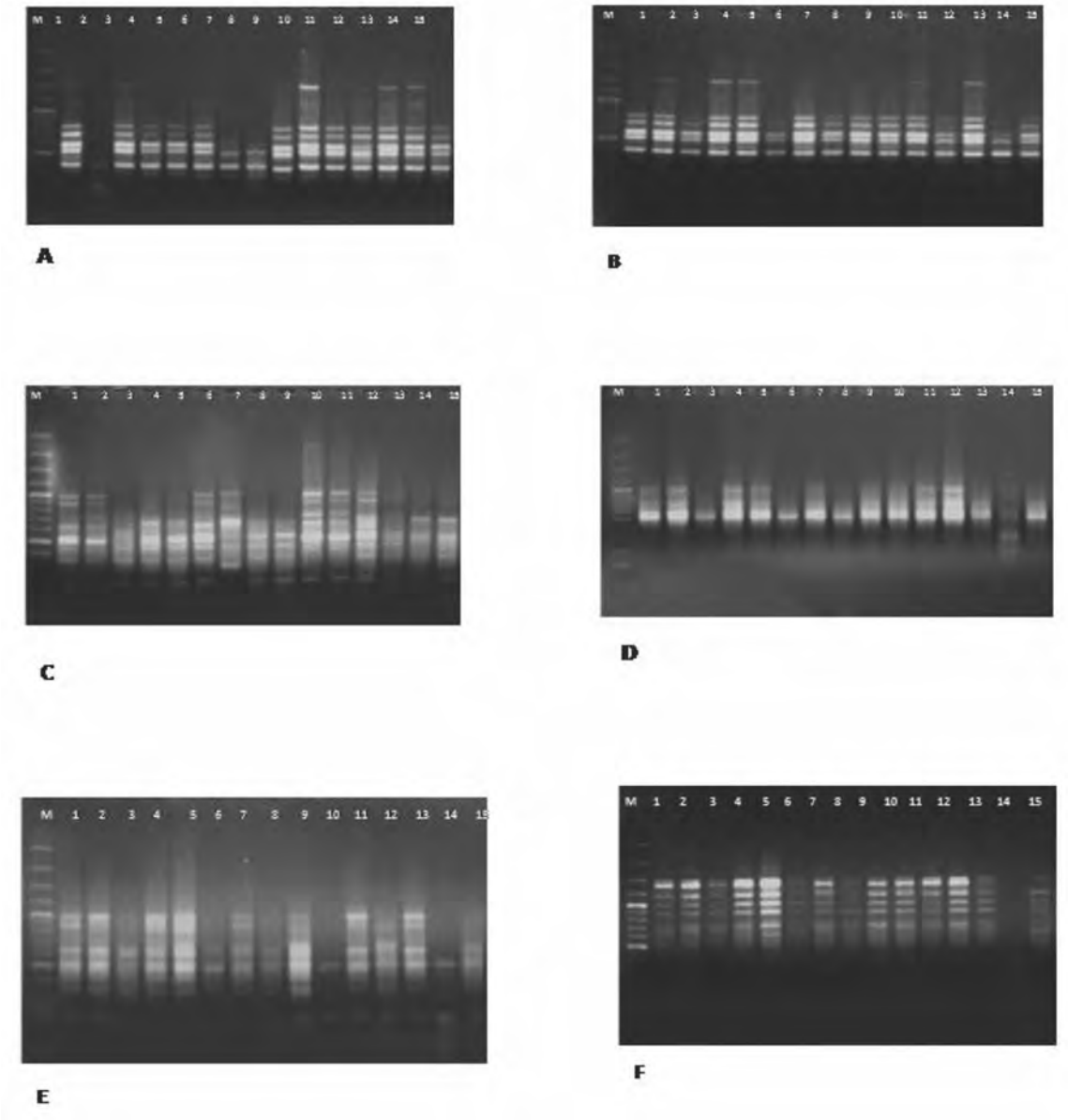

Plate 1. Amplified profile of 15 cotton genotypes with primers (A) OPA 08, (B) OPA 09 and (C) OPA 18. M = ladder, 1= KH 120, 2 = DHY 286 IR, 3 = PH 297 7 1, 4 = KH 121, 5 = NH 572, 6 = L 765, 7 = L 761, 8 = PH 348, 9 = PH 330, 10= PH 44 1 2, 11 = PH 1009, 12 = PH 1024, 13 = NH 545, 14 = Cocker, 15 =MCU 5**.**

polymorphic primers had generated 149 bands. Out of these bands 115 were found polymorphic and 34 were monomorphic. Appreciable amount of polymorphism (*i.e.* 20.00 to 100 %) had generated by these primers confirmed the genetic diversity present among the individual sample. Highly polymorphic primers like OPA 07, OPA 08, OPA 11 and OPA 16 had proved their significance for genetic diversity analysis in cotton.

Using RAPD polymorphism a dendrogram (Fig. 1) was constructed using unweighted pair group method of arithmetic means (UPGMA).

The vertical dashed lines in the

Table 1. Banding pattern generated by polymorphic primers in RAPD analysis of cotton genotypes

Sr. No.	Randomprimer	Total bands	Polymorphic bands	Monomorp hicbands	Polymorphism (%)
1	OPK-01	6	5	1	83.33
2	OPK-03	5	4	1	80.00
3	OPK-04	5	1	4	20.00
4	OPK-05	8	7	1	87.50
5	OPK-07	6	5	1	83.33
6	OPK-08	7	6	1	85.71
7	OPK-09	7	6	1	85.71
8	OPA-05	6	5	1	83.33
9	OPA-06	10	9	1	90.00
10	OPA-07	9	9	0	100.00
11	OPA-08	7	7	0	100.00
12	OPA-09	6	3	3	50.00
13	OPA-10	7	6	1	85.71
14	OPA-11	5	5	0	100.00
15	OPA-12	7	4	3	57.14
16	OPA-13	8	3	5	37.50
17	OPA-15	9	6	3	66.67
18	OPA-16	10	10	0	100.00
19	OPA-17	6	3	3	50.00
20	OPA-18	9	6	3	66.67
21	OPA-20	6	5	1	83.33

dendrogram assigned the genotypes into clusters which correspond well with their centers or sub centers or genetic relationship. In cluster I comprised three cultivars *viz.,* KH 120, KH 121 and L 765 with similarity 0.67, cluster II comprised of three sub clusters, cluster II A comprised three cultivars i.e. DHY 286 IR, MCU 5 and L 761 with similarity coefficient was ranging from 0.75 to 0.77. Cluster II B comprised four cultivars *viz.,* NH 545, NH 572, PH 297 7 1 and PH 348, indicating that they are more closely related which accounted similarity ranges from 0.77 to 0.80. The cluster II C comprised three varieties *viz.,* PH 44 1 2, PH 1009, PH 1024 with similarity value was ranged from 0.79 to 0.82. The cluster III comprised single genotype i.e. Cocker showed 0.66 similarity index. The findings are in accordance with Raina *et al.,* (2001), Archak and Gaikwad (2003), Dongre *et al.,* (2003), Zhang *et al.,* (2004), Dongre *et al.,* (2006), Ebtissam *et al.,* (2007), Zhang *et al.,* (2007), Patil *et al.,* (2007), Sharma *et al.,* (2007), Esmail *et al.,* (2008), Muhammad *et al.,* (2009) and Weian *et al.,* (2009).

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Utilizing heterotic groups of cotton and forming sub groups for further exploitation based on combining ability pattern

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ABSTRACT : Recurrent selection for combing ability was practiced by utilizing heterotic box involving diverse heterotic groups (Robust /stay green *V/s* high RGR) of cotton. A set of two *intra hirsutum* single cross hybrids generated from two elite lines *viz.,* DRGR 24-178 and DRGR-32-100 of high relative growth rate (RGR) group and single elite line from robust line, DSMR-10 and stay green line DSG3-5. These two crosses were advanced to F_4 generation to assess the recombinational variability for combining ability existing among the F_4 lines derived from these crosses. An assessment of combining ability pattern and possibility of development of sub-populations against chosen tester (s) was made. In this method each F_4 line derived from elite lines of each heterotic group was characterized with respect to its combining ability status (pattern) considering the four opposite testers of opposite diverse group. With the help of this information, nature and magnitude of variability for combining ability was assessed individually and based on the derived F_1 performance four sub groups of F_4 lines were formed (A1,A2,A3 and A4....). Results indicated that with respect to reciprocal testers of both populations to enhance the productivity of the hybrids, it was recommended that A1/B1 sub population of high RGR F_4 lines and E1/F1 sub population of robust/stay green F_4 lines could be developed by recombining the best lines of DRGR-24-178 x DRGR-32-100 and DSMR-10 x DSG3-5. At the same time these superior lines could be used to inter cross between these sources as the distance between them was increased which could give a still better cross. By considering the common and diverse testers CT_3 (DH-7225) and DT_4 (DR-8) against high RGR F_4 lines of DRGR-24-178 x DRGR-32-100 and CT_7 (DH-7225) and DT_8 (DRGR-4) against robust/stay green F_4 lines of DSMR-10 x DSG3-5 cross, it was evident that entire group of lines revealed large variability for combining ability to combine with these testers. Between the group of lines tested, the F_4 lines of DRGR-24-178 x DRGR-32-100 was found to be combine much better with these common and diverse testers as compared to the F_4 lines of DSMR-10 x DSG3-5, which was evidenced by the higher frequency of lines falling under the superior category E1, F1, G1 and H1, respectively.

Key words : Combining ability, compact, groups, heterotic, high RGR, robust, stay green, sub grouping

Cotton is an immensely important agricultural crop for the sustainable economy and livelihood of the Indian farming community. It is cultivated in about 33.14 m. ha across the world and in about 11.70 m. ha in the county. India accounts for about 32 per cent of the global cotton area and contributes to 21 per cent of the global cotton produce (37.50 million bales), currently ranking second after China. The domestic consumption of cotton in India is about 25 million bales during 2013-2014. The productivity of cotton in India is about 540 kg/ha, whereas Australia holds highest productivity level (2151 kg/ha) among the major cotton growing nations.

The basic formula on heterosis ($HF_1 = Sdy^2$) explains how performance (heterosis) of hybrid depends on genetic diversity and extent

of dominance existing at different yield influencing loci. It means heterosis can be enhanced either by increasing genetic distance or dominance. It is not possible to manipulate and enhance the degree of dominance and at best we may choose such populations which are differing for the allelic status of such yield influencing loci. If such two base populations are identified which are diverse from each other, it means the plants belonging to the two populations in general differ for the allelic status of yield influencing loci. If each of such chosen populations has inherent variability, this variability can be exploited through selection practised reciprocally to increase the genetic distance further between the populations. In essence, it amounts for widening the genetic distance between the populations (Falconer, 1981).

These principles of population improvement schemes are same in case of both cross and self pollinated crops but the procedure used for enhancing genetic distance (improving combining ability) will change depending upon the type of mating system of crop. Though commercial exploitation of heterosis has taken place at such a revolutionary scale, the hybrid breeding programs are not supported by development of hybrid oriented populations as seen in case of cross pollinated crops. In cross pollinated crops population improvement schemes involving improvement of combining ability as an integral step in evolving hybrids. There is urgent need for implementing such schemes of improving combining ability even in self pollinated crops by crossing the limits of mating system barriers.

In cotton, attempts were made to exploit genetic diversity by forming heterotic groups. Heterotic group is a group of related or unrelated genotypes from the same or different populations, which display similar combining ability and heterotic response when crossed with genotypes from other genetically distinct germplasm groups. By comparison, the term heterotic pattern refers to a specific pair of two heterotic groups, which express high heterosis and consequently high hybrid performance in their cross. Following this line of expectation, an attempt was made at Dharwad to understand in general the complementation pattern of parents contributing to heterosis. The pattern of complementation for plant features has given rise to formation of different heterotic groups like stay green, robust, compact and high Relative Growth Rate (RGR) and their heterotic patterns like stay green x compact, robust x compact, robust x high RGR and stay green x high RGR which in general give potential hybrids.

Development of sub populations with improved combining ability is used as an important step of population improvement schemes in cross pollinated crop like maize. Grouping the lines based on the cross performance of a large set of lines with chosen tester, which could be a known best general combining line and also based on the combining ability status, the genotypes are assigned to the different groups like good, average and poor combiners.

MATERIALS AND METHODS

In the present research, an attempt was made to assess these opposite sets of recombinant lines derived from elite lines of each heterotic groups. The procedure of reciprocal recurrent selection is a method of population improvement practiced by involving diverse base populations (Comstock *et al.,* 1949). After identifying the heterotic box (DRGR-24-178 x DRGR-32-100) x (DSMR-10 x DSG 3-5) during

2009-10. The within group crosses *viz.,* DRGR 24-178 x DRGR 32-100 and DSMR 10 x DSG 3-5 were utilized for developing two diverse base populations for initiating reciprocal selection for combining ability. In both the populations, in the second year, three hundred seeds from each single cross F_1 were advanced to F_2 generation (*kharif*, 2010). In F_2 generation, two hundred good looking plants were selected from each single cross (plant to row progeny) during *kharif*, 2011 and advanced to F_3 generation (Summer, 2012). In F_3 generation, 50 lines were selected randomly and advanced to F_4 generation at Agriculture Research Station, Nipani and Main Agricultural Research Station, Dharwad. The selected 50 F_4 lines of high RGR heterotic group and stay green robust group were utilized for initiating reciprocal selection for combining ability.

The set of 50 high RGR-F_4 lines of DRGR-24-178 x DRGR 32-100 cross and Simultaneously, 50 RSG-F_4 lines of DSMR 10 x DSG-3-5 cross were selected for assessing the variability for combining ability. The set of 50 lines of DRGR 24-178 x DRGR 32-100 were crossed with two reciprocal testers *viz.,* DSMR-10 (RT_1) and DSG 3-5 (RT_2) and similarly 50 lines derived from DSMR 10 x DSG-3-5 were crossed to reciprocal testers *viz.,* DRGR 24-178 (RT_5) and DRGR 32-100 (RT_6). In addition, a new high combiner line (DH 2772) was developed during recent years which combined well with different groups and hence this was used as a common tester against both RGR and RSG-population. For assessing recombination variability for combining ability released in high RGR-F_4 lines, it was proposed to cross them with additional diverse robust lines recently added to the robust group and thus DR 8 (DT_4) was selected as an additional tester against high RGR lines. Similarly, a new high RGR line DRGR 4 (DT_8) was used to assess combining ability of robust/stay green F_4 lines. The efficiency of tester can be judged based on the overall mean of F_1 obtained with that tester and the variability observed among the tester crosses. The F_4 lines derived from DRGR 24-178 x DRGR 32-100 were referred to as population I and those from DSMR 10 x DSG3 5 were referred to as population II. Two hundred hybrids derived from these populations involving F_4 lines with testers were called as derived F_1's (dF_1s). (Fig. 1)

The material pertaining to population I (high RGR heterotic group F_4 lines, testers and its derived F_1s, bench mark crosses (original non parental crosses) and commercial *Bt* checks (Kanaka, Chiranjeevi and JK Durga) were raised in one block (Table 1) and that pertaining to population II (against stay green/robust F_4 lines) were raised in the adjoining block, during *Kharif*, 2013 at ARS, Belvatigi. (Table 2). The derived F_1s were planted in a randomized block design with two replications/each population. A spacing of 90 cm between rows and 60 cm between the plants within a row was followed, taking two rows/ replication and a row length of 4.80 m. Fertilizers at recommended doses were applied and other cultural practices were carried out at regular intervals. The plant protection measures were taken up at appropriate time to control pests and diseases.

Grouping of lines based on combining ability : The overall mean of all the crosses with the tester can be worked out and four sub groups of tester cross performance of combining ability can be made as stated below. This sub grouping of can be done separately for each of the testers with which they are crossed.

Based on the mean (of all the crosses), the crosses were divided into four classes and given the ranks as '1' (> mean + SD), '2' (mean to mean + SD), '3' (mean to mean – SD) and '4' (<

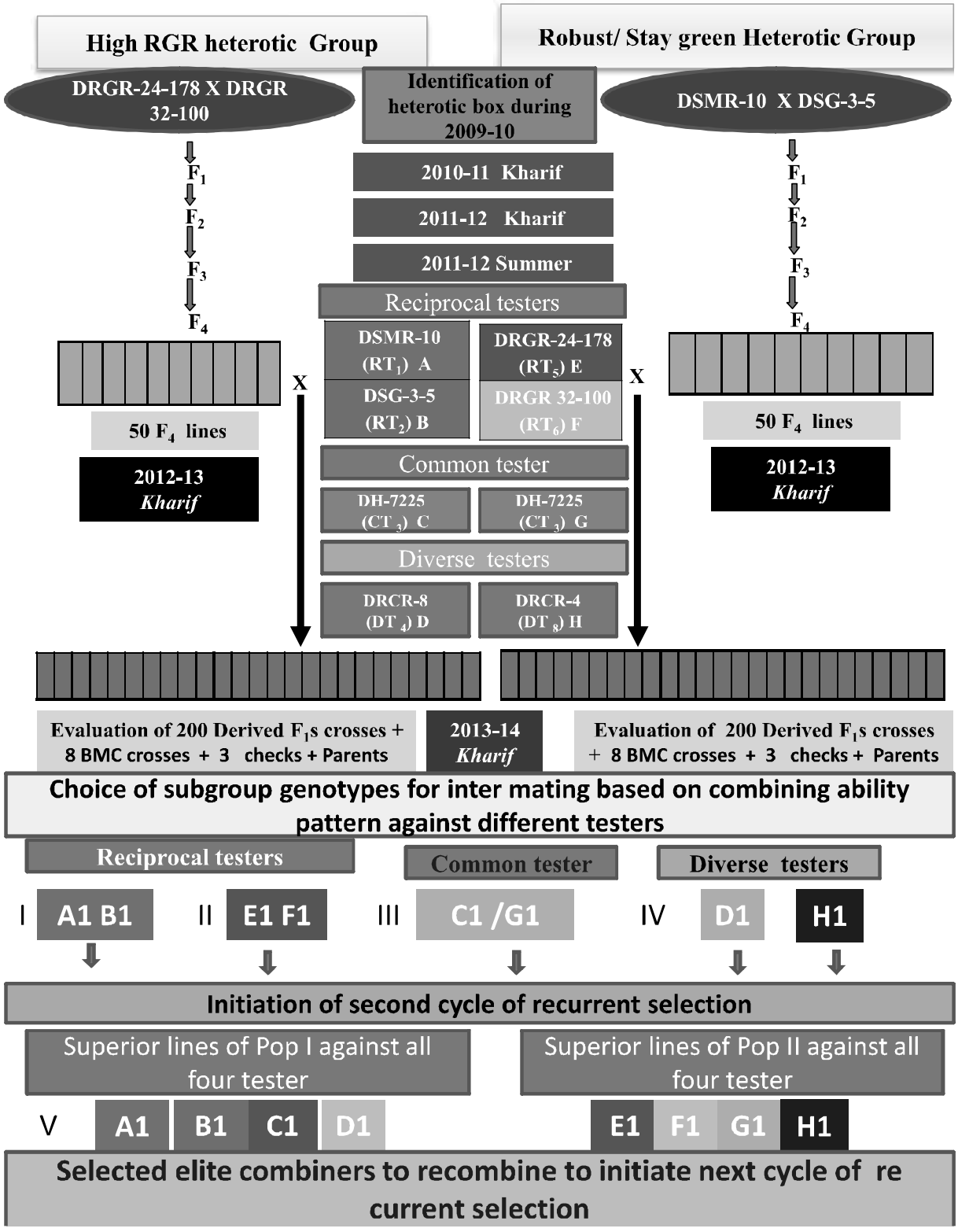
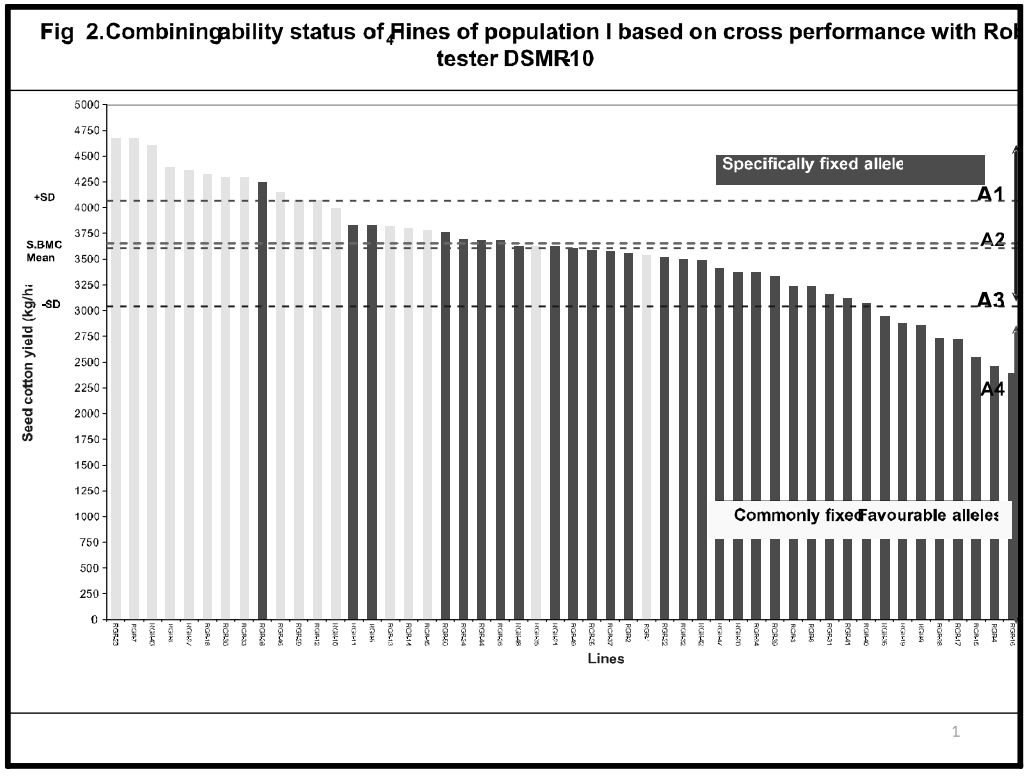


Fig. 1. Practicing one cycle of reciprocal selection for combining ability in cotton and forming sub groups based on combing ability pattern

mean – SD) as suggested by Patil (1995). Each line was characterized with respect to its combining ability status (pattern) against the testers. With the help of this information, the nature and magnitude of variability for combining ability was assessed.

The information obtained from the cross between the F_4 lines of both the population with respect to its combining ability status (pattern) to corresponding testers. By following the method of determining the combining ability pattern given above, each line of two populations were categorized and compared with the other lines. The A1 subgroup consist of the best combiner against the tester utilized. It means that these have possibly maximum number of favourable alleles and these favourable alleles compliment perfectly with the favourable alleles of the tester. From A1 sub group can be involved in developing the A1 sub population (Al-SP) for improving combining ability in general and combining ability with this tester in particular. This also means that the sub population can give which are most likely to combine very well with other from the population to which the tester represents.

In another form of graphical representation shown in Figures. Two testers can be chosen for representation, one on each axis. Here it has been shown by utilizing the derived F_1 performance of from pop I and II with the reciprocal , common and diverse testers inducted in the study. This form of representation helps in identifying the falling genotypes in 16 sub groups formed in each graph. The falling genotypes in AlBl similarly C1Dl, E1F1 and G1F1 subgroups represent the most potential combiners against the tester combinations concerned. In the same graph the mean performance of each sub group against the testers is depicted. Further, the scoring patterns of the are given and the top five crosses are



marked from examining the combining ability pattern of the lines involved in the best crosses.

It is possible to choose revealing high combining ability against a particular chosen tester. Such chosen can be involved in developing sub population for improving combining ability against the chosen tester (s). For example, if the objective is to improve combining ability against RT_1, the representing A1 sub group can be utilized in developing A1 sub population which will recombine and accumulate favourable alleles for giving superior performance against RT_1.

RESULTS AND DISCUSSION

In this study, two complimentary populations (Pop I and II) which are developed from two diverse single cross F_1s, are crossed with each other by using opposite parents as a tester to identify those of pop I capable of combine well with pop II and *vice versa*. Among the F_4 lines of DRGR 24 187 x DRGR 32 100 (pop I), ten lines belongs to this group *viz.*, RGR 23, RGR 7, RGR 43, RGR 8, RGR 27, RGR 18, RGR 30, RGR 33 RGR 28 and RGR 46 belonged to higher combiner category (A1) against the opposite tester DSMR 10 (RT_1) /A (Fig. 2). The ten F_4 lines *viz.*, RGR 9, RGR 50, RGR 8, RGR 10, RGR 15, RGR 49, RGR 13, RGR 17, RGR 1 and RGR 12 fell under higher combiner category (B1) against the opposite population DSG3 5 (RT_2) /B (Fig. 3). In case of population II, F_4 lines of DSMR 10 x DSG3 5 *viz.*, Seven F_4 lines RSG 31, RSG 17, RSG 45, RSG 19, RSG 25, RSG 36 RSG 23 and RSG 29 belonged to higher combiner category (E1) against the opposite tester DRGR 24 178 (RT_5) /E (Fig. 4). Whereas, 12 F_4 lines *viz.,* RSG 23, RSG 48, RSG 18, RSG 38, RSG 29, RSG 25, RSG 24, RSG 49, RSG 36, RSG 27, RSG 17 and RGR 50 fell under higher combiner category (F1) against the

Table 1. Parental genotypes used for study on exploitation of identified heterotic box through reciprocal selection combining ability involving high RGR F_4 lines of DRGR 24 178 x DRGR 32 100.

	50 F_4 lines of Population I (DRGR 24 178 x DRGR 32 100)		
Sl. No.	High RGR F_4 lines	Sl. No.	High RGR F_4 lines
1	RGR 1	26	RGR 26
2	RGR 2	27	RGR 27
3	RGR 3	28	RGR 28
4	RGR 4	29	RGR 29
5	RGR 5	30	RGR 30
6	RGR 6	31	RGR 31
7	RGR 7	32	RGR 32
8	RGR 8	33	RGR 33
9	RGR 9	34	RGR 34
10	RGR 10	35	RGR 35
11	RGR 11	36	RGR 36
12	RGR 12	37	RGR 37
13	RGR 13	38	RGR 38
14	RGR 14	39	RGR 39
15	RGR 15	40	RGR 40
16	RGR 16	41	RGR 41
17	RGR 17	42	RGR 42
18	RGR 18	43	RGR 43
19	RGR 19	44	RGR 44
20	RGR 20	45	RGR 45
21	RGR 21	46	RGR 46
22	RGR 22	47	RGR 47
23	RGR 23	48	RGR 48
24	RGR 24	49	RGR 49
25	RGR 25	50	RGR 50

Testers

1 DSMR 10 (Reciprocal Tester) (RT_1)
2 DSG 3 5 (Reciprocal Tester) (RT_2)
3 DH 7225 (Common Tester) (CT_3) DR 8
4 (Diverse Tester) (DT_4)

Checks (Popular Bt hybrids)

1 Kanaka *Bt*
2 Chiranjeevi *Bt*
3 JK Durga *Bt*

Bench mark crosses

1 DRGR 24 178 x DSMR10
2 DRGR 32 100 x DSMR10
3 DRGR 24 178 x DSG3 5
4 DRGR 32 100 x DSG3 5
5 DRGR 24 178 x DH 7225
6 DRGR 32 100 x DH 7225
7 DRGR 24 178 x DR 8
8 DRGR 32 100 x DR 8

Table 2. Parental genotypes used for the study on exploitation of identified heterotic box through reciprocal selection for combining ability involving robust/stay green (RSG) F_4 lines of DSMR 10 x DSG3 5

50 F_4 lines of Population II (DSMR 10 x DSG3 5)			
Sl. No.	Robust stay green lines	Sl. No.	Robust stay green lines
1	RSG 1	26	RSG 26
2	RSG 2	27	RSG 27
3	RSG 3	28	RSG 28
4	RSG 4	29	RSG 29
5	RSG 5	30	RSG 30
6	RSG 6	31	RSG 31
7	RSG 7	32	RSG 32
8	RSG 8	33	RSG 33
9	RSG 9	34	RSG 34
10	RSG 10	35	RSG 35
11	RSG 11	36	RSG 36
12	RSG 12	37	RSG 37
13	RSG 13	38	RSG 38
14	RSG 14	39	RSG 39
15	RSG 15	40	RSG 40
16	RSG 16	41	RSG 41
17	RSG 17	42	RSG 42
18	RSG 18	43	RSG 43
19	RSG 19	44	RSG 44

Testers

1 DRGR 24 178 (Reciprocal tester) (RT_5)
2 DRGR 32 100 (Reciprocal tester) (RT_6)
3 DH 7225 (Common tester) (CT_7)
4 DRGR 4 (Diverse tester) (DT_8)

Checks (Popular Bt hybrids)

1 Kanaka *Bt*
2 Chiranjeevi *Bt*
3 JK Durga *Bt*

Bench mark crosses

1 DRGR 24 178 x DSMR10
2 DRGR 32 100 x DSMR10
3 DRGR 24 178 x DSG3 5
4 DRGR 32 100 x DSG3 5
5 DSMR10 x DH 7225
6 DSG3 5 x DH 7225
7 DSMR10 x DRGR 4
8 DSG3 5 x DRGR 4

opposite population DRGR 32 100 (RT_6) /F (Fig 5). The superior F_4 lines of the two populations identified in this study are genetically more diverse than the original base populations. In the next phase of work, these superior lines from two populations need to be crossed between them to assess the performance of the crosses based on the lines improved through reciprocal selection for combining ability.

The number of lines selected for synthesizing a subpopulation may vary depending on the differences existing among the top order combiners. If just top two are distinctly superior than the rest and hence among the good combiner if top two are chosen they can be crossed to develop F_2 population. The segregating generations of this cross can be evaluated for combining ability by using the same tester. A pedigree method of breeding for combining ability can be followed in which selection is practiced for combining ability, the trait which is measured based on tester cross performance with the concerned tester.

Based on graphical presentation given for common tester, it was evident that lines of two populations revealed a larger variability for ability to combine with the tester. The F_4 lines of DRGR 24 187 x DRGR 32 100 combined better with the DH 7335 (CT_3) / C as evidenced by the eight dF_1 of this figured in the top rank C1 *viz.*, RGR 8, RGR 13, RGR 12, RGR 20, RGR 28, RGR 4, RGR 18 and RGR 49 (Fig. 6) and similarly 13 dF_1 *viz.,* RSG 18, RSG 23, RSG 6, RSG 25, RSG 27, RSG 47, RSG 31, RSG 7 RSG 28, RSG 9, RSG 33, RSG 11 and RSG 37 in case of the F_4 lines of DSMR 10 x DSG 3 5 of this figured in the top rank D_1 (Fig. 7) .

Based on graphical presentation given for diverse tester it was evident that the lines of two populations revealed a larger variability for ability to combine with the tester. The F_4 lines

Fig. 3: Combining ability status of lines of population I based on cross performance with Stay tester DSG-25

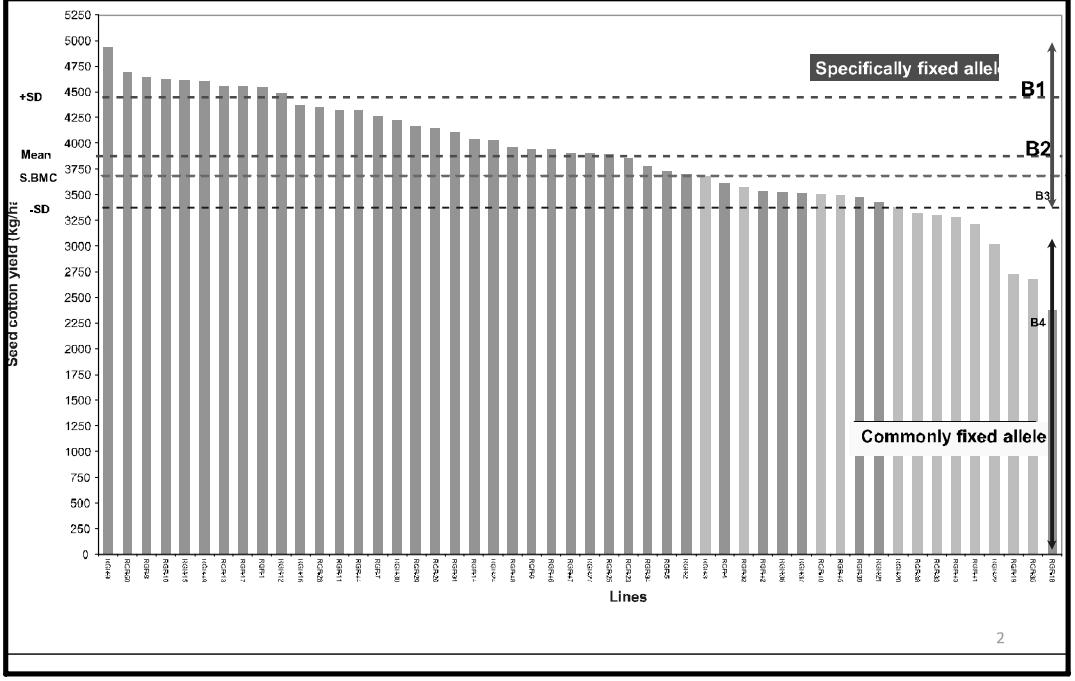
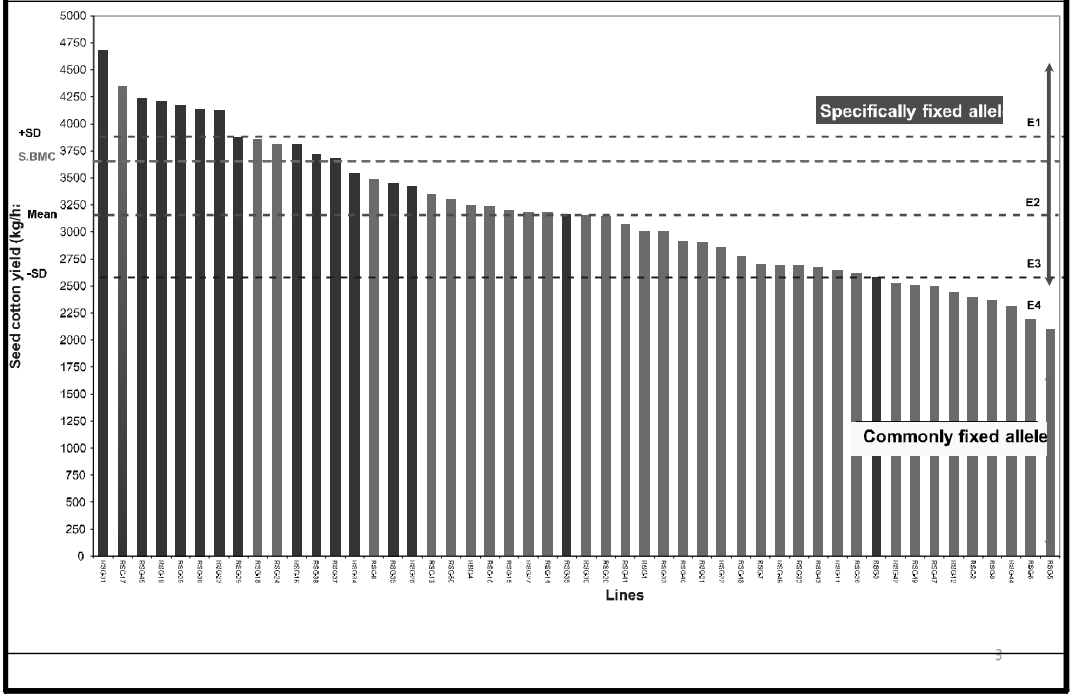
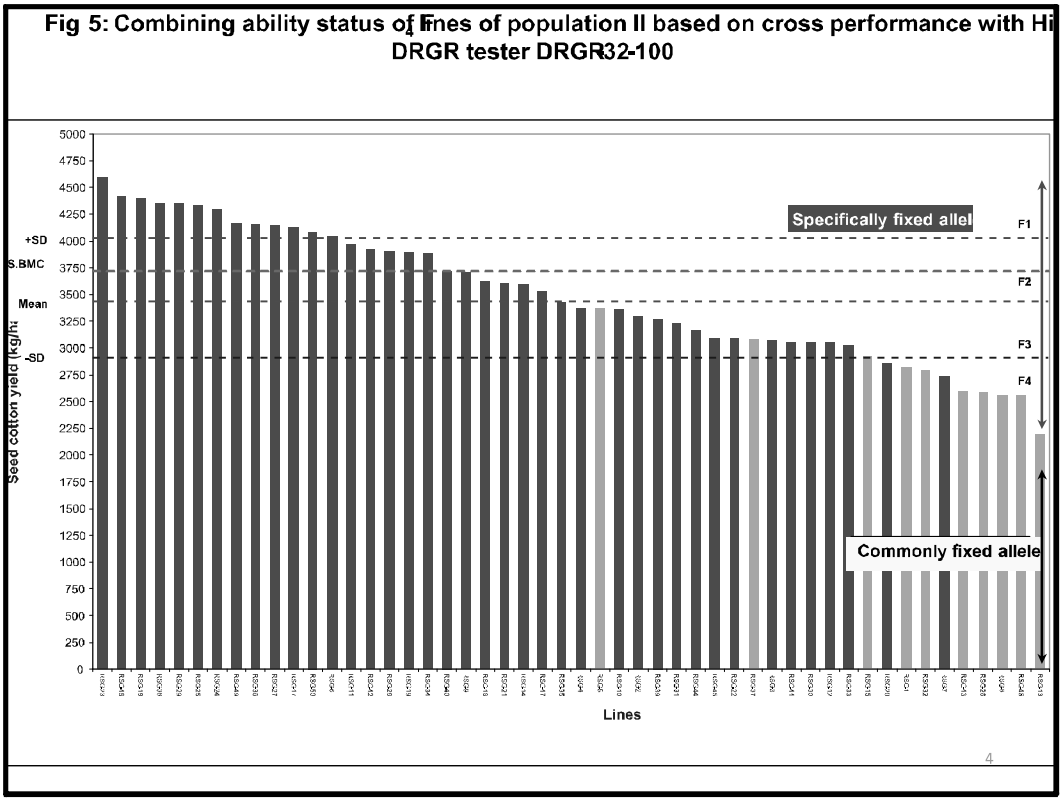


Fig 4: Combining ability status of lines of population II based on cross performance with High tester DRGR-24-178





of DRGR 24 187 x DRGR 32 100 combined better with the DR 8 (DT_4) / G as evidenced by the higher frequency of crosses falling under the category G 1 (Fig. 8) *viz.*, RGR 30, RGR 23, RGR 34, RGR 22, RGR 13, RGR 47 and RGR 50. In case of population II, F_4 lines of DSMR 10 x DSG3 5 *viz.*, RGR 45, RGR 20, RGR 23, RGR 37, RGR 17, RGR 18, RGR 9, RGR 22, RGR 25, RGR 48 and RGR 6 found in higher combiner category (H1) against the diverse tester DRGR 4 (DT_8) /H (Fig 9).

In another form of graphical representation shown in Figures. Two testers

Table 3. Distribution of transgressive segregants of high RGR F_4 lines of DRGR 24 178 x DRGR 32 100 against reciprocal tester

	Positive transgressive segregants above the mean value	
Sl. No.	Lines	Classes
1	RGR 8	A1B1
2	RGR 50, RGR 10, RGR 12, RGR 13	A1B2
3	RGR 28, RGR 30, RGR 07, RGR 27, RGR 23	A2B1
4	RGR 46, RGR 29, RGR 11, RGR 14, RGR 24, RGR 44, RGR 26, RGR 48	A2B2
Negative transgressive segregants		
5	RGR 19	A4B4

Table 4. Distribution of transgressive segregants of robust stay green (RSG) F_4 lines of DSMR 10 x DSG3 5 against reciprocal tester

	Positive transgressive segregants above the mean value	
Sl. No.	Lines	Classes
1	RSG 23, RSG 29, RSG 18, RSG 25, RSG 36, RSG 17,	E1F1
2	RSG 19	E1F2
3	RSG 38, RSG 24,	E2F1
4	RSG 16, RSG 34, RSG 28, RSG 50	E2F2
Negative transgressive segregants		
5		E4F4

Fig. 6: Combining ability status of lines of population I based on cross performance with Com tester DH-7225

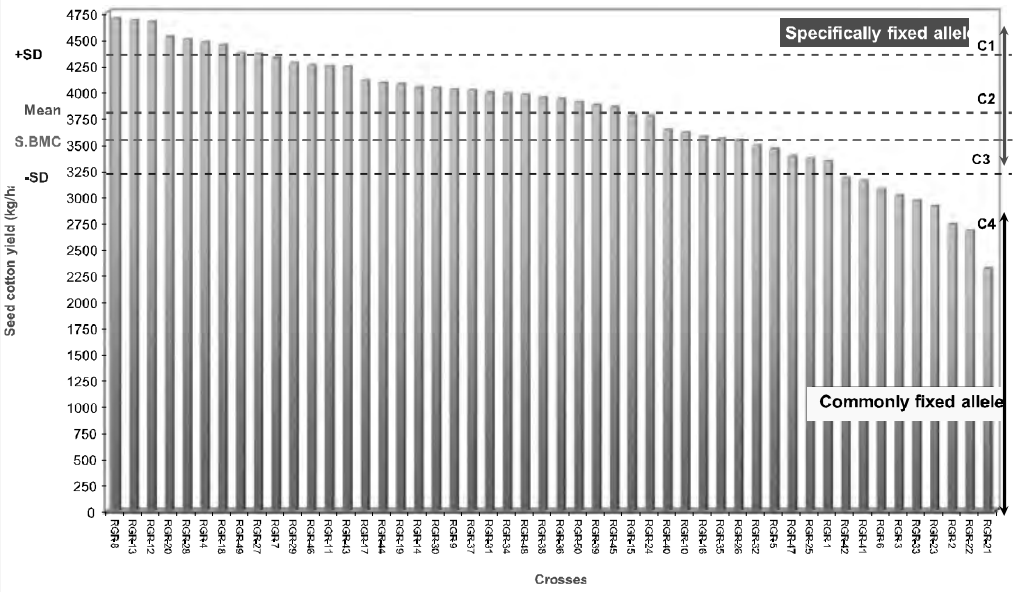


Fig 7 : Combining ability status of lines of population I based on cross performance with Div tester DRCR-8

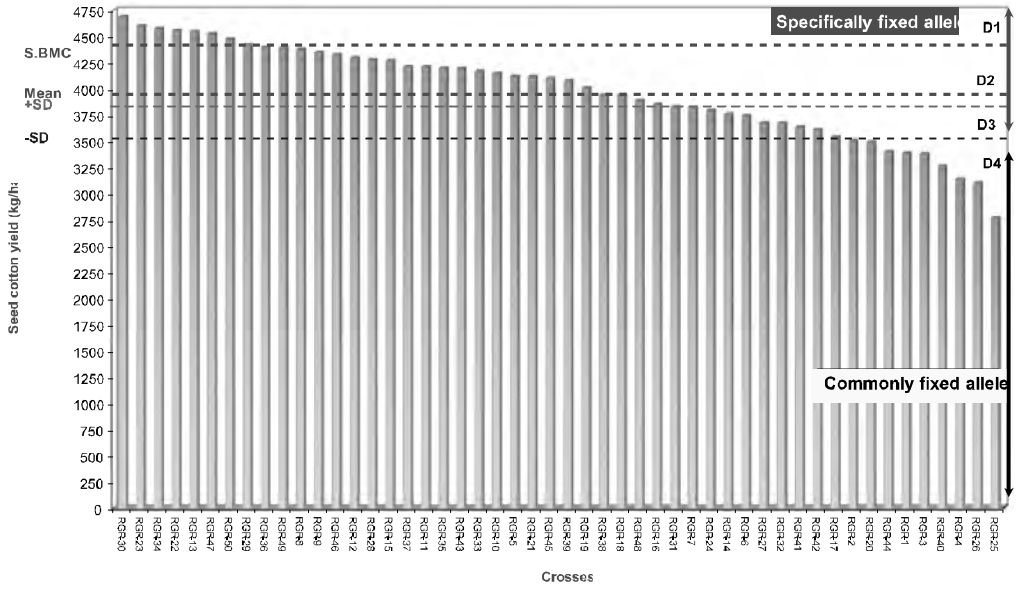


Fig. 8 : Combining ability status of lines of population II based on cross performance with Con tester DH-7225

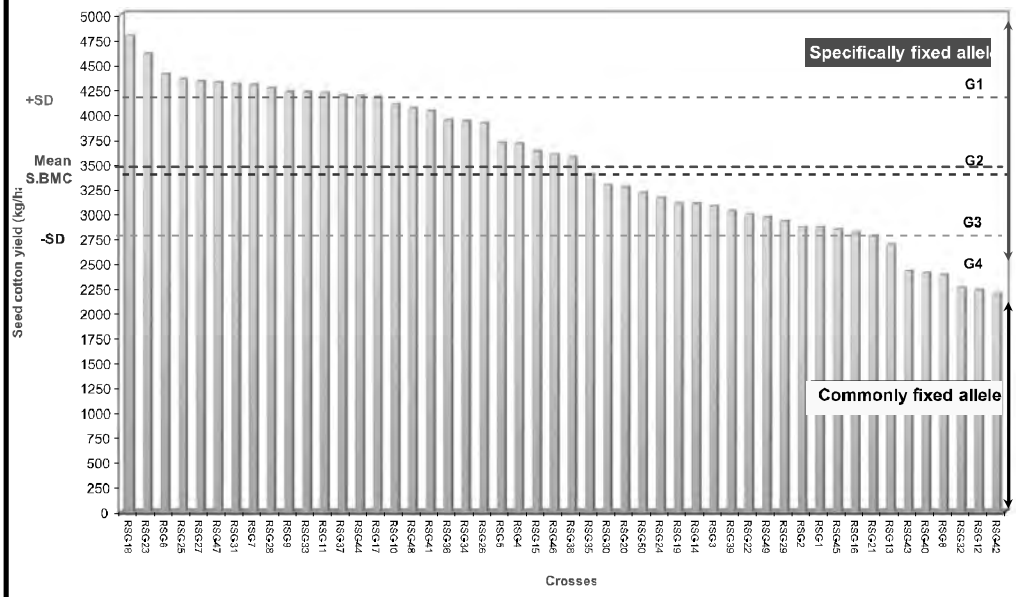


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Fig. 9 : Combining ability status of lines of population II based on cross performance with div RGR tester DRCR-4

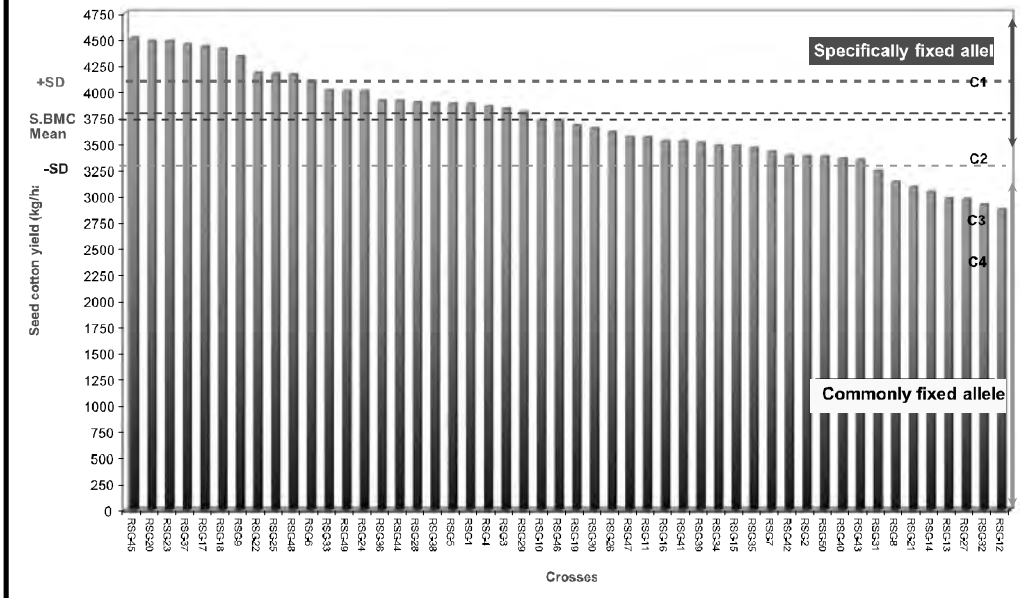

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Fig. 10 Sub-grouping of F_4 lines of population I based on cross performance against two Robust/Stay Green reciprocal testers (DSMR-10 and DSG3-5)

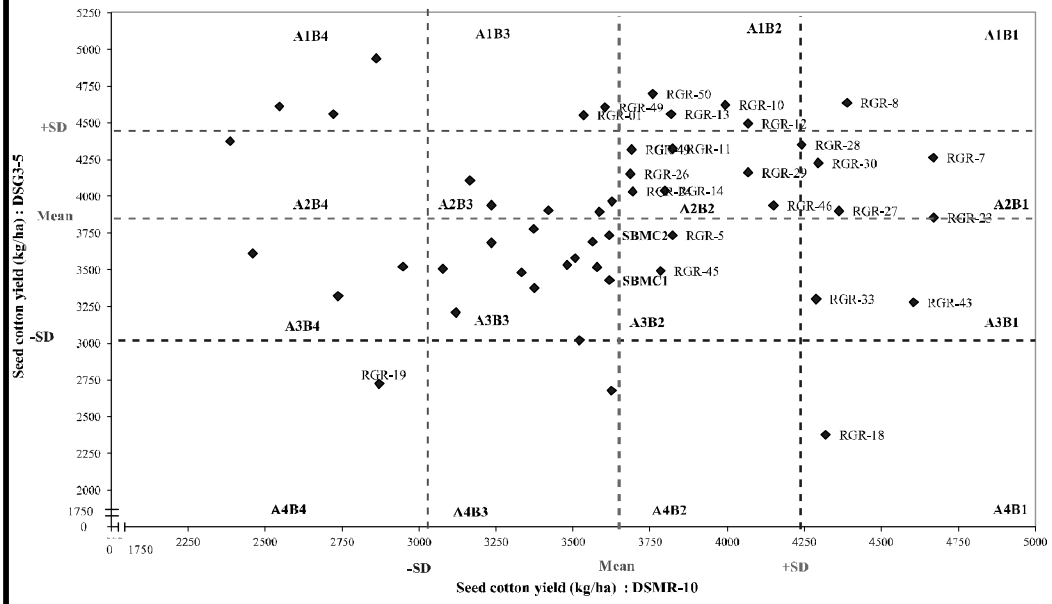

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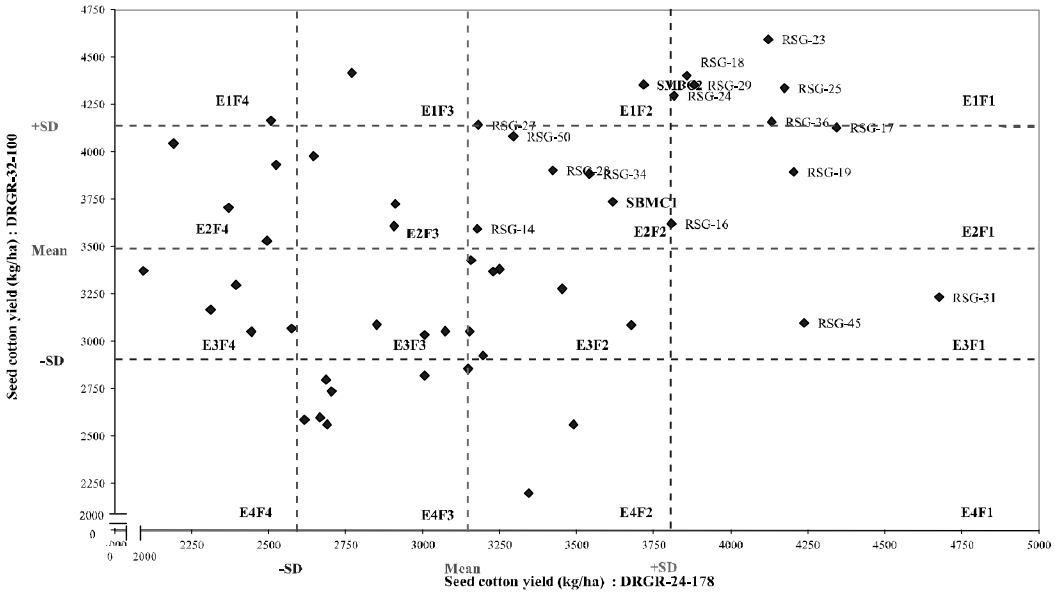
Fig. 11 : Sub-grouping of F_4 lines of population II based on cross performance against two High RGR reciprocal testers (DRGR-24-178 and DRGR-32-100)



10

can be chosen for representation, one on each axis. Here it has been shown by utilizing the F_1 performance of from pop I and II with the reciprocal and common and diverse testers inducted in the study. This form of representation helps in identifying the falling genotypes in 16 sub groups formed in each graph. The falling genotypes in AlBl similarly C1Dl, E1F1 and G1F1 subgroups represent the most potential combiners against the tester combinations concerned. In the same graph the mean performance of each sub group against the testers is depicted. Further, the scoring patterns of the are given and the top five crosses are marked from examining the combining ability pattern of the lines involved in the best crosses.

It is possible to choose revealing high combining ability against a particular chosen tester. Such chosen can be involved in developing sub population for improving combining ability against the chosen tester (s). For example, if the objective is to improve combining ability against RT_1, the representing A1 sub group can be utilized in developing A1 sub population which will recombine and accumulate favourable alleles for giving superior performance against RT_1.

Based on the graphical presentation given for tester DSMR 10 (RT_1) /A and DSG3 5 (RT_2) / B (Fig 10), A1 is a class of lines which are more diverse to the reciprocal tester DSMR 10 (RT_1) and B1 is the class of lines which are more diverse to the another reciprocal tester DSG3 5 (RT_2). They have accumulated highest number of specifically fixed favourable allele in those respective F_4 lines. whereas, A1B1 is the class of lines which are simultaneously diverse to the both the reciprocal tester. It is evident that line *viz.,* RGR 8 was belong to class of A1B1 revealed large variability for ability to combine with these two reciprocal testers (Table 3). Similarly in the graphical representation of tester DRGR 24 178 (RT_5) /E and DRGR 32 100 (RT_6) / F (Fig. 11). E1F1 is the class of lines which are simultaneously diverse to the both the reciprocal tester. It is evident that lines RSG 23, RSG 29, RSG 18, RSG 25, RSG 36, RSG 17 were belong to class of E1F1 revealed large variability for ability to combine with these two reciprocal tester (Table 4). Whereas the graphical representation of common tester DH 7225 (CT_3) /C and diverse tester DR 8 (DT_4) / D against high RGR F_4 lines

Table 5. Distribution of transgressive segregants of high RGR F_4 lines of DRGR 24 178 x DRGR 32 100 against common (DH 7225) and diverse tester (DR 8)

	Positive transgressive segregants above the mean value	
Sl. No.	Lines	Classes
1	RGR 13	C1D1
2	RGR 30, RGR 34, RGR 50	C1D2
3	RGR 8, RGR 49, RGR 12, RGR 28, RGR 12, RGR 49	C2D1
4	RGR 46, RGR 29, RGR 11, RGR 19, RGR 9, RGR 37, RGR 36, RGR 39, RGR 45	C2D2
Negative transgressive segregants		
5	RGR 3 and RGR 2	C4D4

Table 6. Distribution of transgressive segregants of robust stay green (RSG) F_4 lines of DSMR 10 x DSG3 5 against common (DH 7225) and diverse tester (DR 8)

	Positive transgressive segregants above the mean value	
Sl. No.	**Lines**	**Classes**
1	RSG 18, RSG 23, RSG 25	G1H1
2	RSG 37, RSG 17, RSG 9	G1H2
3	RSG 6, RSG 28	G2H1
4	RSG 33, RSG 44, RSG 10, RSG 48, RSG 36, RSG 5, RSG 4, RSG 46, RSG 38	G2H2
Negative transgressive segregants		
5	RSG 21, RSG 13, RSG 8, RSG 32, RSG 12	G4H4

Fig. 12 : Sub-grouping of F_4 lines of population I based on cross performance against common (DH-7225) and diverse (DRCR-8)testers

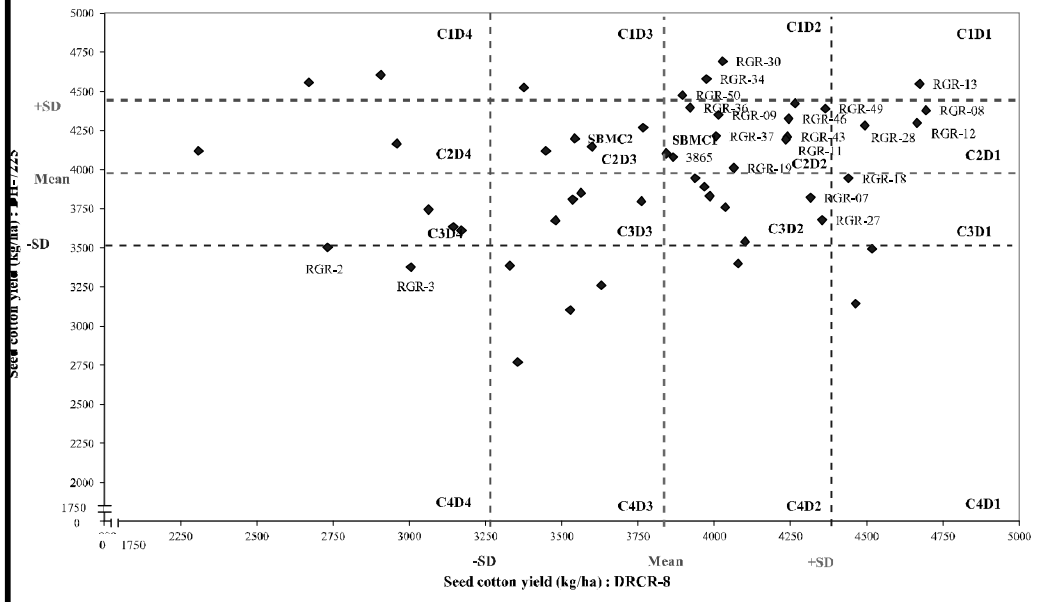


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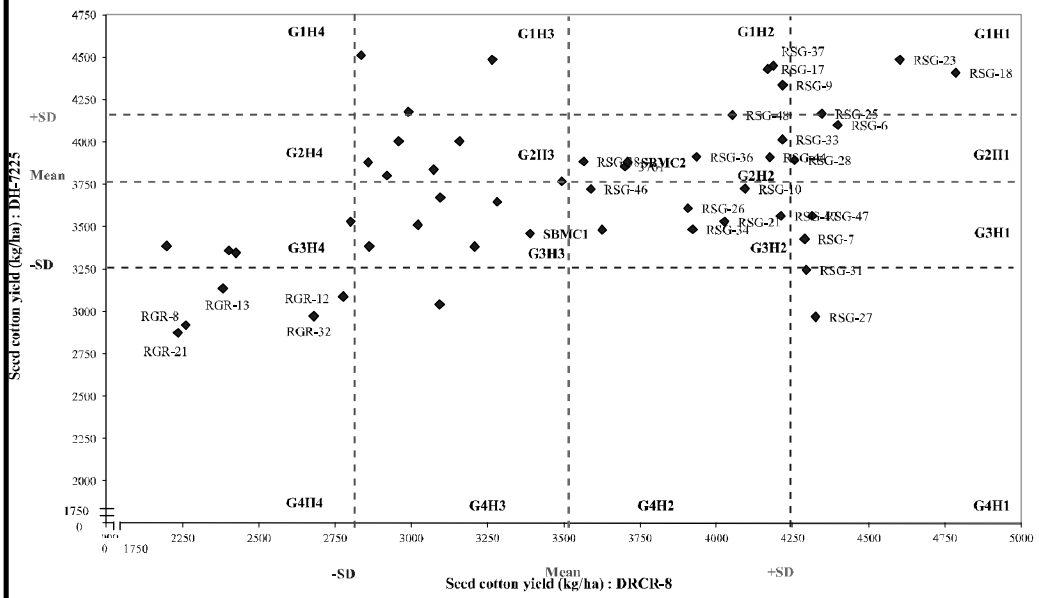
Fig. 13: Sub-grouping of F_4 lines of population II based on cross performance against common (DH-7225) and diverse (DRCR-4) testers



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(Fig 11) grouped in C1D1 class of lines which are simultaneously diverse to both common and diverse tester. It is evident that line RGR 13 was belong to class of C1D1 revealed large variability for ability to combine with these common and diverse tester (Table 5). This suggest that this first class line can recombined with other potential lines to initiate second phase of creating recombinational variability for combining ability.

With respect to the population II, the graphical representation of common tester DH 7225 (CT_7) /G and diverse tester DRGR 4 (DT_8) / H against robust and stay green F_4 lines (Fig. 13) grouped in G1D1 class of lines which are simultaneously diverse to the both common and diverse tester. It is evident that lines RSG 18, RSG 23 and RSG 25 were belong to class of G1H1 revealed large variability for ability to combine with these common and diverse tester (Table 6). This also suggested that these first class lines can recombined to initiate second phase of creating recombinational variability for combining ability.

CONCLUSION

In routine population improvement scheme meant for improving combining ability there are three steps namely a) Selfing b) Evaluation of the elite combiners c) Recombining the elite combiner. The principle of population improvement for combining ability has been extended in this study to a self pollinated crops. Analysis of variance of results revealed what are good combiner F_4 lines of the population for individual testers and also pairs of testers. The elite combiners whose derived F_1's revealing seed cotton yield more than Mean + 1 Standard Deviation are identified and it is likely that lines belonging to this elite group may differ with respect to identity of dominant favorable allele present in them. Hence the next course of action should be recombine elite combiners to develop a new population to initiate the next cycle of reciprocal recurrent selection against a single testers or combination of testers.

The elite high combiner lines identified from opposite populations are expected to be genetically diverse. Hence the crosses obtained from these elite combiners are expected to be potential. The elite sub populations give scope for further enhancement of genetic distance between the groups and paves way for the next phase of improvement in hybrid performance. With this presumption the elite high RGR F_4 lines of DRGR 24 178 x DRGR 24 178 *viz.*, RGR 23, RGR 7, RGR 43, RGR 8, RGR 27, RGR 9, RGR 50, RGR 10, RGR 15 and RGR 49 and the robust stay green elite F_4 lines of DSMR 10X DSG3 5 opposite population *viz.,* RSG 31, RSG 17, RSG 45, RSG 19, RSG 25, RSG 23, RSG 48, RSG 18, RSG 38 and RSG 29 were selected on the basis of derived F_1s performance and the ten elite combiners each from the opposite groups will be crossed in line x tester fashion to get new improved derived F_1s.

The trangressive segregants distributed in population I and II were classified in to positive and negative transgressive segregants based on the sub grouping of F_4 lines with respect to reciprocal tester, common and diverse testers respectively where positive transgressive segregants are showing positive gca effect with the opposite parents hence they combined well the both parents. There are array of lines which combine well with single parent and is due to the recombination of allele which favouring P_1 or P_2 alone. Similarly negative transgressive segregants are those which are showing

significant negative *gca* effect with the opposite parent.

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Genetical and biochemical basis of cotton leaf curl disease in upland cotton

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ABSTRACT : The present investigation comprising six generations (Parents, F_1, F_2, BC_1 and BC_2) of four crosses in cotton *viz.,* H1098-I x B 59-1678, H 1117 x HS6, H 1098-I x H1117 and B 59-1678 x HS6 was conducted for studying the inheritance of cotton leaf curl virus disease and estimating the gene effects for the yield and its component traits during *kharif,* 2013. The inheritance of cotton leaf curl virus disease indicated the duplicate dominant epistasis(15:1). Generation mean analysis revealed significant differences for all traits indicating thereby the presence of non-allelic interactions. In some of the cases, the non-significance of chi-square value indicated the fitness for additive-dominance model. Additive component was significant for most of the characters and even as preponderant in magnitude over the dominance component. Either all or any of the three types of epistatic interactions (i, j and l) were significant for most of the cases and generally it is the "i" type of interaction which is more frequently prevailing for most of the traits studied over the crosses. Additive x additive type of interaction was recorded for plant height, boll number, boll weight, GOT, seed index and seed cotton yield. Duplicate type of interaction was apparent for plant height, boll number, boll weight, GOT and lint index. Among biochemical parameters, sugar content was higher in susceptible parents than resistant ones. Also, it decreased at 90 DAS when the disease incidence was higher and further increase at 120 DAS. Phenol, tannin and gossypol content, the secondary metabolites were higher in resistant parents as compared to susceptible parents. Concentration was more at 90 DAS which decrease at 120 DAS. Correlation matrix indicated that cotton leaf curl virus disease grading was significant positively correlated with the sugar content and negatively correlated with phenol, tannin, gossypol, protein and cellulose.

Cotton as a crop as well as commodity plays an important role in the agrarian and industrial activity of the nation and has a unique place in the economy of our country. Cotton is grown in tropical and sub-tropical regions of more than 80 countries world over while leading cotton producing countries are China, India, USA, and Pakistan (Meyer *et al.*, 2013).

Cotton is the most important *kharif* cash crop of north India. In Haryana only *G. hirsutum* and *G. arboreum* species are grown. More than 90 per cent area of cotton is under *G. hirsutum*. Main reasons for low productivity of cotton in India attributed to the high incidence of insect pests and diseases caused by fungal, bacterial and viral pathogens. Of these viral diseases alone or in combination with other factors are quite destructive and are limiting factor for the cotton cultivation resulting significant loss in seed cotton yield.

The diseases caused by geminiviruses are of considerable concern. Among various geminiviruses cotton leaf curl virus disease CLCuD is the most devastating disease in cotton. In India, cotton leaf curl virus disease was first reported in American cotton (*G hirsutum*) in Sriganganagar area of Rajasthan state during 1993 (Ajmera,1994) and during 1994 it appeared in Haryana and Punjab (Rishi and Chauhan,1994;Singh *et al.*,1994) states on

hirsutum cotton and posed a major threat to its cultivation in northern India (Varma *et al.*,1995). The disease has appeared in an epidemic form during 1997 in the Rajasthan affecting an area of 0.1 million hectares (Anonymous,1998).

Use of chemicals in controlling the whitefly (the vector of this virus) is costly and not so effective. Moreover, it may be hazardous to men and environment. Extensive uses of pesticides have also caused damage to soil quality and fertility (Dinham, 1993). Therefore, development of a resistant variety to this disease is the most effective, long term, less expensive and safe method to fight against this disease and to enhance the productivity of cotton. Research efforts to develop resistant varieties/ hybrids through conventional/ biotechnological approaches along with cultural and management practices are in progress for effectively controlling this disease.

Nature has provided cotton with traits like okra leaf type, gossypol glands and trichomes which confer non-preference to the insect pest infestation. The information on various biochemical parameters imparting resistance to cotton leaf curl virus disease will help for identification of resistant varieties based on biochemical attributes and same can be used as early selection criterion for screening germplasm and other breeding material.

In order to achieve this goal, an understanding of mode of inheritance of cotton leaf curl virus disease and seed cotton yield along with other biochemical parameters (responsible for resistance to cotton leaf curl disease) is necessary for proper choice of breeding procedures.

MATERIALS AND METHODS

The present investigation was conducted at Cotton Research Area, Department of Genetics and Plant Breeding, CCS Haryana Agricultural University, Hisar during *kharif,* 2013. Four parents two resistant (H 1098I and H 1117) and two susceptible (B 59-1678 and HS 6) to cotton leaf curl virus disease were chosen to generate the experimental material. These four parents were used to develop four crosses, H 1098-I x B 59-1678 (R x S), H 1117 x HS 6 (R x S), H 1098-I x H1117(R x R) and B 59-1678 x HS 6 (S x S). These crosses were designated as cross I, cross II, cross III and cross IV, respectively and finally six generations were generated *i.e.* P_1, P_2, F_1, F_2, BC_1 and BC_2. The experimental material comprised of six generations *i.e.* parents (P_1 and P_2), F_1, F_2 and back crosses (B_1 and B_2) of four crosses was grown in a compact family block design with three replications. There was a single row of non segregating generations (P_1, P_2 and F_1), 20 rows of F_2, and 8 rows of each BC_1 and BC_2 generations. The length of each row was being 6 m with a spacing 67.5 x 30 cm. In order to build up heavy inoculums pressure one row of highly susceptible line (HS 6) was planted at the periphery of the experimental area.

Observation on CLCuD was recorded under field condition in each replication on all the plants of each of the non –segregating generations (P_1, P_2 and F_1), backcross generations and the F_2 generation. Disease was scored on 0 -5 grade depending upon the response to the cotton leaf curl virus disease CLCuD.

$$\text{Per cent disease incidence} = \frac{\text{Number of diseased plants}}{\text{Total number of plants}} \times 100$$

0= Immune; complete absence of symptoms, per cent disease incidence = 0

1= Highly resistant; very minute thickening of veins, per cent disease incidence = 0 – 10

2= Resistant; thickening of small group of veins, per cent disease incidence=10–20

3= Susceptible; severe vein thickening and leaf curling developed at the top of the plant, per cent disease incidence=20- 40

4= Moderately susceptible; severe vein thickening and leaf curling developed on the half of the plant canopy, per cent disease incidence = 40 - 70

5= Highly susceptible; severe vein thickening, leaf curling, enation and full stunting of plant, per cent disease incidence = 70 – 100

Five competitive plants from each row of non – segregating generations and 30 plants from F_2 generation and 10 plants from each of backcrosses were chosen at random for recording observations on the following economic characters: days to first flower, plant height, no. of bolls / plant, boll weight, ginning outturn, seed index, lint index and seed cotton yield/plant

Biochemical study was carried out in biochemistry laboratory of Cotton Section, Department of Genetics and Plant Breeding, CCS HAU, Hisar. Total phenols, total sugars, tannins, crude proteins, total gossypol and structural carbohydrates were analyzed using their standard methods. The healthy leaves as well as diseased leaves of all four crosses were taken at three stages of plant growth i.e. vegetative stages (60 DAS), reproductive stage (90 DAS) and maturity stage (120 DAS).

Statistical Analysis

The Chi – Square test of goodness of fit

$$\chi^2 = i = \sum_{1}^{n} \frac{(Oi - Ei)^2}{Ei}$$

Whereas;

O_i = observed frequency of i^{th} class

E_i = expected frequency of i^{th} class

For testing the significance, the c^2 tabulated values were seen at n-1 d.f.

Biometrical analysis for Estimation of Gene Effects : The "t" statistical test was applied to test the differences between parental genotypes for the studied characters before considering the biometrical analysis. The gene effects were estimated by employing generation mean analysis of Mather (1967), Hayman and Mather (1955) and Jinks and Jones (1958).

RESULTS AND DISCUSSION

The incidence of cotton leaf curl virus disease during the experimental year *i.e.* 2013-2014 was very severe under field condition particularly nearby Hisar areas including CCS HAU, cotton research area. During this year no variety/ strain was observed completely immune to this disease. Even in highly resistant strains only few plants showed immunity. The F_1s *viz.*, H 1098-I x B 59-1678 and Cross H 1117 x HS 6 had resistance to CLCuD indicated that resistance is a dominant trait. The expression of resistance in both (R x S) crosses revealed

Table 1. Inheritance of CLCuD in upland cotton

Parent/ generation	Number of plants			Expected ratio	a”^{2} calculated	P value
	Screened	Resistant	Susceptible			
Cross I (R x S): H 1098-I x B 59-1678						
P_1	54	54	0			
P_2	56	0	56			
F_1	61	61	0			
F_2	364	348	16	15:1	2.27	0.131
BC_1	218	218	0			
BC_2	243	173	70	3:1	1.77	0.183
Cross II(R x S):Cross H 1117 x HS 6						
P_1	61	61	0			
P_2	57	0	57			
F_1	59	59	0			
F_2	358	331	27	15:1	0.74	0.389
BC_1	221	221	0			
BC_2	231	168	63	3:1	0.575	0.448
Cross III (R x R): H 1098-I x H1117						
P_1	54	54	0			
P_2	59	59	0			
F_1	58	58	0			
F_2	347	347	0			
BC_1	236	236	0			
BC_2	221	221	0			
Cross IV(S x S): 59-1678 x HS 6						
P_1	59	0	59			
P_2	59	0	59			
F_1	56	0	56			
F_2	358	0	358			
BC_1	230	0	230			
BC_2	235	0	235			

that there was no cytoplasmic inheritance for the expression of susceptibility to CLCuD. The dominance nature of resistance over susceptibility was further confirmed by backcrosses and F_2s. The pattern of segregation in F2 gave a good fit to 15 resistant: 1 susceptible (Table 1) indicating the duplicate type of gene action. Disease was expressed in those plants which had recessive genes at both the loci. Duplicate type of gene interaction for CLCuD was further confirmed by a good fit of 3 resistant: 1 susceptible ratio of backcross with susceptible parents.

The breeding for cotton leaf curl disease resistance has been achieved through the assemblage of minor genes by recurrent selection and according to Azhar *et al.*(2010) resistance depends on major genes (dominant genes) which may lose quickly because of the evolution of pathogen for these genes. The F_1 of crosses between highly susceptible S 12, highly resistant LRA 5166 varieties were found all virus free plants and their F_2 was close to 3:1 ratios which exhibit the presence of a single gene for the inheritance of resistance against CLCuD reported by Rehman *et al.,* (2005). Ahuja *et al.*, 2006 reported 4 types of segregation patterns in the F_2 generations. A good fit for 15 (resistant):1

(susceptible), 13 (resistant):3 (susceptible), 9 (resistant):7 (susceptible) ratios indicated digenic control of the trait with duplicate dominant, dominant inhibitory, and duplicate recessive epistasis, respectively. Three gene controls with triplicate dominant epistasis was obtained in one of the crosses.

In F2 generation of cross H 1098-I x H1117 (R x R), all the plant were resistant resistant to CLCuD indicated that genes involved in the resistance of CLCuD were present at the same locus in both the parents. Hence no segregation pattern was observed in F2 and backcrosses. In cross 59-1678 x HS 6 (S x S), all the plants in F1, F2 and backcross generations were susceptible to CLCuD. The disease reaction in all generations was similar to the reaction of parents suggesting that there was no complimentary interaction between the genes for susceptibility in both the susceptible parents.

Estimation of epistasis: The joint scaling test of Cavalli (1952) has indicated the adequacy of simple additive-dominance model for days to first flower in cross I, II and III, 100 seed weight (cross III), lint index (cross III) and seed cotton yield in crosses I, III and IV. Additive effects for seed cotton yield and its attributing characters were also reported by Kaushik and Kapoor, (2006), Abbas *et al.*, (2008), Ali *et al.*, (2009), Lu and Myers, (2011) and Iqbal *et al.*, (2013). This suggested that the additive gene effects played important role in the inheritance of all these attributes and simple selection would be adequate to improve such characters.

The characters under study which could not be explained on simple additive-dominance model as tested through scaling tests were analyzed on digenic epistatic model of Hayman (1958). The estimates of mean (m), additive (d), dominance (h), additive x additive (i), additive x dominance (j) and dominance x dominance (l) were estimated from six generations i.e. P_1, P_2, F_1, F_2, BC_1 and BC_2.

For those characters, where digenic model has been found as adequate, largely the characters have been observed where there has been preponderance of both 'additive' and 'dominance' components and among epistatic components mostly 'i' type (additive x additive) and 'l' type (dominance x dominance) epistatis contributed significantly towards the gene effects. Moreover, in some other situations either 'i-type' epistatis alone or 'i-type' epistatis in combination with 'j-type' epistatis or all the three types of epistatis were found significantly contributing to the gene effects. It is interesting to note that 'j' type of epistasis alone has been reported only in few cases viz. seed index (cross IV), lint index (cross II and IV). Preponderance of additive x additive (i-type) epistasis or gene interaction suggested that such traits in the population maybe improved through random mating of the selected desirable plants followed by selection. This approach will lead to the exploitation of additive (d); additive x additive (i-type) of gene effects and interactions in the populations. The high frequency of occurrence of dominance (h) and dominance x dominance (l-type) gene effects and interactions may paradoxically suggest the exploitation of heterosis in cotton. However, a close examination for the sign of 'h' and 'l' type of epistatis reveal that magnitude of the two if found in opposite direction than contribution to the phenotypic mean imply thereby antagonistic effects in heterosis expression and it has been termed as 'duplicate' type of epistasis which may be explained on the basis of fact that majority of the parents involved in the cross were selections towards a single optimum phenotype and as such it is this selection for optimum type that has

Table 2(a). Generation mean analysis for yield attributing traits in different crosses of upland cotton

Characters	Parameter	Cross I (H1098-I x B59-1678)	Cross II (H 1117 x HS 6)	Cross III (H 1098-I x H 1117)	Cross IV (B59-1678 x HS 6)
Days to first flower	**Joint scaling test (three parameter model)**				
	m	55.77**±0.36	56.87**±0.55	55.63**±-0.42	54.95**±0.39
	d	1.04**±0.33	0.18±0.54	0.73±0.41	-0.47±0.37
	h	-0.01±0.69	0.34±1.02	0.20±0.79	0.36±0.76
	÷²(df=3)	**4.98**	**3.03**	**0.63**	**8.39****
	Six parameter model				
	m	56.02**±0.30	57.10**±0.33	55.88** ±0.31	55.65**±0.31
	d	-0.93±0.61	-0.60±0.91	0.07± 0.34	0.000±0.60
	h	-0.85±1.89	-2.00±2.49	-1.28±2.12	4.02±1.93
	i	-0.22±1.74	-2.53±2.26	-1.48±1.95	-4.35±1.75
	j	0.20±1.48	-0.93±2.29	-0.46±1.79	-1.86±1.55
	l	-3.44±3.13	7.06±4.41	2.48±3.62	7.82±3.17
	Type of epistasis	-	-	-	-
Plant height	**Joint scaling test (three parameter model)**				
	m	131.94**±1.43	138.27**±1.68	136.58**±1.34	129.22**±1.28
	d	-21.28**±1.46	-6.02**±1.69	-5.08**±1.37	13.91**±1.31
	h	-1.28±2.38	-6.29±3.21	-2.28±2.33	-7.22**±2.17
	÷²(df=3)	**27.17****	**146.57****	**442.82****	**272.76****
	Six parameter model				
	m	128.88**±2.05	117.81**±2.16	107.36**±2.49	102.51**±2.15
	d	15.60**±3.73	3.33±4.58	22.56**±5.86	-11.80*±5.09
	h	-20.75±11.35	45.72**±13.04	2.60±15.57	6.52±13.51
	i	-20.08±11.09	40.75**±12.60	-2.33±15.39	8.75±13.33
	j	-8.66±8.16	-0.60±9.89	42.20**±12.07	1.53±10.54
	l	64.62**±17.70	45.04*±21.36	170.66**±25.92	115.31**±22.54
	Type of epistasis	Duplicate	Complimentary	-	-
Boll number	**Joint scaling test (three parameter model)**				
	m	13.09**±0.36	14.57**±0.56	14.39**±0.43	11.98**±0.48
	d	1.99**±0.36	3.50**±0.51	0.33±0.45	-0.42±0.45
	h	6.16**±0.85	4.05**±1.19	4.73**±0.79	8.29**±1.06
	÷²(df=3)	**10.76****	**13.68****	**8.63****	**13.67****
	Six parameter model				
	m	16.34**±0.57	16.30**±0.54	16.20**±0.46	15.46**±0.55
	d	2.30*±1.09	6.66**±1.05	-0.86±1.52	1.86±0.96
	h	8.45*±3.32	8.63*±3.41	14.63**±3.66	12.90**±3.26
	i	4.15±3.16	3.06±3.02	10.00*±3.57	6.66*±2.94
	j	0.66±2.32	8.33**±2.41	-1.26±3.20	3.80±2.18
	l	-14.02*±5.36	-1.40±5.69	-18.33*±6.58	-15.93**±5.25
	Type of epistasis	Duplicate	-	Duplicate	Duplicate
Boll weight	**Joint scaling test (three parameter model)**				
	m	3.53**±0.03	33.82**±0.17	3.43**±0.04	3.00**±0.04
	d	-0.39**±0.03	-0.13±0.16	-0.42**±0.04	0.05±0.04
	h	-0.23**±0.07	-2.73**±0.37	-0.32**±0.08	-0.19*±0.07
	÷²(df=3)	**72.19****	**31.88****	**17.63****	**8.54****
	Six parameter model				
	m	3.39**±0.08	2.98**±0.03	3.32**±0.04	3.08**±0.31
	d	0.31**±0.08	-0.12*±0.05	0.55**±0.10	-0.28±0.13
	h	-1.05**±0.38	-0.72**±0.22	-1.02**±0.28	-1.07±1.29
	i	-0.99**±0.37	-0.32±0.18	-0.72**±0.26	-0.91±1.29
	j	-0.09±0.19	0.10±0.16	0.41±0.22	-0.53±0.29
	l	2.93**±0.50	1.71**±0.35	1.54**±0.47	1.30±1.38
	Type of epistasis	Duplicate	Duplicate	Duplicate	

Table 2(b). Generation mean analysis for yield attributing traits in different crosses of upland cotton

Characters	Parameter	Cross I (H1098-I x B59-1678)	Cross II (H 1117 x HS 6)	Cross III (H 1098-I x H 1117)	Cross IV (B59-1678 x HS 6)
Ginning Outturn	**Joint scaling test (three parameter model)**				
	m	34.3**±0.10	34.44**±0.14	33.97**±0.23	33.82**±0.17
	d	-0.99**±0.10	-0.56**±0.13	-0.05±0.22	-0.13±0.16
	h	-0.44±0.22	-1.30**±0.29	0.63±0.41	-2.73**±0.37
	÷²(df=3)	**23.64****	**12.99****	**17.33****	**31.88****
	Six parameter model				
	m	34.25**±0.16	33.67**±0.20	43.85**±0.17	31.77**±0.17
	d	2.20**±0.34	1.24**±0.24	0.71*±0.28	-0.06±0.41
	h	-2.08*±0.98	-0.17±1.00	3.45*±1.41	1.70±1.19
	i	-1.80±0.95	1.38±0.94	1.72±0.91	3.20**±1.10
	j	2.74**±0.71	1.82**±0.59	2.82±2.17	-0.43±0.91
	l	3.63*±1.59	2.95*±1.43	-1.72±2.55	-0.01±2.03
	Type of epistasis	Duplicate	-	-	-
Seed index	**Joint scaling test (three parameter model)**				
	m	7.45**±0.04	7.27**±0.06	7.34**±0.05	7.17**±0.05
	d	-0.77**±0.04	0.48**±0.06	-0.59**±0.05	0.60**±0.04
	h	0.36**±0.09	0.52**±0.10	-0.22*±0.09	0.48**±0.10
	÷²(df=3)	**20.65****	**31.68****	**4.05**	**15.23****
	Six parameter model				
	m	7.89**±0.07	7.41**±0.07	7.29**±0.06	8.29**±0.71
	d	0.58**±0.10	-0.25±0.13	0.72**±0.13	-0.15±0.13
	h	-1.09**±0.37	0.22±0.40	-0.26±0.38	-2.79±2.85
	i	-1.42**±0.36	-0.31±0.39	-0.02±0.36	-3.18±2.85
	j	-0.45±1.76	0.63*±0.29	0.28±0.29	1.01**±0.29
	l	1.76**±0.55	1.96**±0.63	-0.46±0.62	2.66±2.90
	Type of epistasis	Duplicate	-	-	-
Lint index	**Joint scaling test (three parameter model)**				
	m	3.93**±0.03	3.82**±0.03	3.77**±0.06	3.68**±0.03
	d	-0.59**±0.03	0.14**±0.03	-0.35**±0.06	0.27**±0.03
	h	0.08±0.06	-0.04±0.06	-0.03±0.11	-0.05±0.07
	÷²(df=3)	**17.49****	**24.23****	**6.21**	**7.11**
	Six parameter model				
	m	4.12**±0.05	3.77**±0.04	3.73**±0.03	3.85**±0.32
	d	0.68**±0.08	0.08±0.08	0.49**±0.08	-0.08±0.08
	h	-0.92**±0.27	0.05±0.26	0.42±0.27	-0.98±1.32
	i	-1.02**±0.26	0.05±0.25	0.30±0.22	-0.93±1.31
	j	0.22±0.18	0.65**±0.18	0.53±0.32	0.46*± 0.19
	l	1.49**±0.42	0.49±0.40	-0.48±0.48	1.18± 1.36
	Type of epistasis	Duplicate	-	-	-
Seed cotton yield	**Joint scaling test (three parameter model)**				
	m	39.27**±1.40	40.18**±1.55	41.55**±1.39	27.91**±1.20
	d	-11.97**±1.32	-9.73**± 1.33	-3.97*±1.44	0.16±1.17
	h	12.98**±3.05	-0.52±3.28	8.51**±2.54	19.67**±2.69
	÷²(df=3)	**0.41**	**24.65****	**4.39**	**7.17**
	Six parameter model				
	m	45.39**±1.52	38.92**±1.11	44.65**±1.21	37.29**±3.54
	d	13.26**±2.94	16.78**±2.30	6.54±3.43	-0.13±3.24
	h	16.18±9.34	17.66*±8.01	25.42**±8.80	28.64±15.90
	i	2.28±8.48	6.18±6.40	17.07*±8.40	13.18±15.58
	j	3.26±6.60	21.22**±5.67	5.70±7.57	0.22±6.96
	l	-0.40±15.41	20.42±14.07	-31.02*±15.46	-36.42±20.23
	Type of epistasis	-	-	-	-

favoured the duplicate but not the complementary interaction (Mather, 1967). Hence, it is difficult to improve the populations in the presence of duplicate type of epistatis.

Biochemical studies : In recent years, it is becoming increasingly evident that several natural and induced defense mechanisms operate in host plants against different diseases. Sometimes, the host plant is induced to synthesize these compounds on infection. The content of different biochemical constituents viz., sugar content, total phenol, tannin, gossypol, protein and cellulose were estimated at three different growth stages in leaf samples i.e. 60 days after sowing, 90 days after sowing and 120 days after sowing (DAS) in all the four crosses. The results obtained are described as under:-

Table 3. Different biochemical parameters among resistant and susceptible parents at 60 DAS

Parents	Sugar (%)	Phenol (%)	Tannin (%)	Gossypol (%)
H 1098-i	1.43	1.33	0.62	0.50
H 1117	1.20	1.46	0.57	0.48
B 59-1678	2.24	0.80	0.41	0.34
HS 6	3.26	0.78	0.39	0.27
S.E.(m)	0.08	0.04	0.01	0.02
C.D. (p=0.05)	0.25	0.10	0.04	0.05

Sugars acts as precursor for synthesis of phenolics, phytoalexins, lignin and cellulose which play an important role in defense mechanism of plants against invading pathogens (Klement and Goodman, 1967). In the present investigation, sugar content was less in resistant parents (H 1117 and H 1098—I) as compared to that of susceptible parents (B 59 – 1678 and HS 6).

Among major secondary metabolites of different plants, phenols stand out as most important component in imparting resistance to several plant diseases. Higher concentration causes an instant lethal action by a general tanning effect while, lower concentration causes gradual effect on the cellular constituent of the parasite (Dasgupta, 1988). The total phenol content under both the situations (healthy and infected) increased from 60 to 90 DAS. The rate of increase in the total phenol content in response to the CLCuD infection was more in infected plants as compared to healthy ones. This result is in agreement with the findings of Borkar and Verma (1991) in case of cotton against Bacterial blight, Chakrabarty *et al.,* (2002) in case of cotton against Grey mildew, (Govindappa *et al.,* 2008) in case of cotton against bacterial blight.

Tannins are astringent, bitter-tasting plant polyphenols that bind and precipitate proteins. Tannins are considered to be the most important secondary plant compound involved in plant defense against insects and disease (Swain, 1979). In the present investigation, tannin content was higher in resistant plants as compared to susceptible ones. The tannin content under both the situations (healthy and diseased) increased significantly from 60 to 90 DAS and further decreased from 90 to 120 DAS. The rate of decrease in the tannin content in response to the CLCuV infection was more in infected plants as compared to healthy ones. This result is in agreement with the findings of Beniwal *et al.*, (2006) and Acharya and Singh (2008).

Cotton produces a number of toxic terpenoid aldehyde (TA) compounds contained in epidermal glands that help protect the plant from pests and diseases. Gossypol is one of the major TA and its content varied genotypically and with plant age. The gossypol content was 44.69 and

Table 4. Correlation matrix among different biochemical parameters of crosses I: H1098-i x B59-1678 at 60 DAS

Cross I 60 days	CLCuD grading	Sugar	Phenol	Tannin	Gossypol	Protein	Cellulose
CLCuD grading	1.000	0.913**	-0.927**	-0.964**	-0.898**	-0.897**	-0.927**
Sugar		1.000	-0.852**	-0.890**	-0.796**	-0.796**	-0.790**
Phenol			1.000	0.891**	0.767**	0.767**	0.904**
Tannin				1.000	0.866**	0.865**	0.876**
Gossypol					1.000	0.849**	0.836**
Protein						1.000	0.836**
Cellulose							1.000

79.12 per cent was lower in susceptible parents than resistant parents in crosses H 1098-I x B 59-1678 and H 1117 x HS 6 respectively. Also, the gossypol content in both susceptible and resistant plants was increased significantly from 60 to 90 DAS and then decreased from 90 to 120 DAS.

Correlation matrix:

The correlation matrix among different biochemical parameters revealed that with the CLCuD (Cotton Leaf Curl Virus Disease) grading (disease scoring 0-5) and sugar content showed positive significant correlation (Table 4) while other biochemical parameters viz. phenol, tannin, gossypol, protein and cellulose showed significant negative correlation. CLCuD grading did not showed significant correlation with oil content. The same trend was observed in all the four crosses and at different stages of growth.

SUMMARY

The inheritance of cotton leaf curl virus disease indicated the duplicate dominant epistasis(15:1). No complementary gene interaction was observed in cross with susceptible parents for CLCuD resistance.

Scaling tests revealed that additive-dominance model was fit for the characters, namely, Days to first flower in crosses (H 1098-I x B 59- 1678), (H1117 x HS 6), (H1098-I x H 1117), seed index (cross H1098-I x H 1117), lint index in crosses (H1098-I x H 1117and B 59- 1678 x HS 6) and seed cotton yield in crosses (H 1098-I x B 59- 1678, H1098-I x H 1117 and B 59- 1678 x HS 6).All the three types (i, j and 1) or either of them of epistatic effects were significant for most of the cases wherein additive x additive (i) type of interaction was reported for plant height, boll number, boll weight, GOT, seed index and seed cotton yield.

These different biochemical parameters viz. tannin, phenol, gossypol provides defense mechanism to plants so have higher content in resistant and tolerant plants while susceptible have lower values. Also, sugar act as substrate for insects that's why susceptible plants have higher sugar content as compared to tolerant ones. However, there is no threshold level studied so far as it depends upon environmental conditions, genotypes, maturity stages and also varies from plant to plant.

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Study of molecular variability of *Alternaria* spp on *Bt* cotton

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ABSTRACT: RAPD analysis of *Alternaria* spp on *Bt* cotton revealed that the maximum genetic similarity of 93 per cent was between Gabbur (A_7) and Sunkeshwarhal (A_8) isolates, whereas the least similarity (40 per cent) was observed between Hattigudoor (A_9) and Yaragatti (A_{13}) isolates. The data was differentiated the isolates of *Alternaria* spp. into two major clusters *i.e.*, A cluster and B cluster. Cluster A was further sub-grouped in to 2 sub-clusters *viz.*, A1 encompassing Bijapur (A_{11}) isolate and A2 encompassing Yaragatti (A_{13}) and Bagalkot (A_{16}) isolates. Cluster B was further sub grouped in to 2 sub-clusters *viz.*, B1 and B2. The sub cluster B1 comprised of Kannolli (A_{11}) isolate. The sub cluster B2 comprised of the remaining isolates *i.e.* Dediapada (A_{17}), Ulligeri (A_{14}), Hattigudoor (A_9), Hanumanamatti (A_{12}), Kalmala (A_6), Javalgeri (A_5), Dharwad (A_4), Dharwad Farm (A_3), Annigeri (A_2), Sunkeshwarhal (A_8), Gabbur (A_7), Hirebagevadi (A_{15}) and UAS Dharwad (A_1) isolates. In B2 sub cluster maximum similarity was found between Gabbur (A_7) and Sunkeshwarhal (A_8) isolates as well as Annigeri (A_2) and Dharwad Farm (A_3) isolates. From the results it was clear that, all the isolates belonging to one geographical location have come in the same cluster, reflecting the fact that the variation is dependent on geographical locations. The PCR amplification and sequencing of ITS rDNA region of fungus was best molecular tool for identification of *Alternaria* spp. This is the first time of a sequence of *Alternaria dianthi* and two *Alternaria* spp. isolated from *Bt* cotton are to be publishing in this national symposium.

Key words: *Alternaria* spp, *Bt* cotton, Isolates, molecular variability

Cotton is one of the most ancient and important commercial crops next only to food grains and is the principal raw material for a flourishing textile industry. Currently, *Gossypium* includes 50 species, four of which are cultivated, forty four are wild diploids, and two are wild tetraploids (Percival and Kohel, 1990). Out of the four cultivated species, *Gossypium hirsutum* L. and *Gossypium barbadense* L. commonly called as new world cottons are tetraploids (2n = 4x = 52). Whereas, *G. herbaceum* L. and *G. arboreum* L. are diploids (2n = 2x = 26) and are commonly called as old world cottons.

There has also been a manifold improvement in production, productivity and quality with virtual increase in area. India now produces around 371.20 lakh bales of cotton ranging from short staple to extra long staple from an area of 121.91 lakh ha with productivity of 481.23 kg/ha (Anonymus, 2012). In Karnataka, the area under cotton cultivation is 5.49 lakh ha with a production of 13.10 lakh bales and an average productivity of 405.65 kg/ha (Anonymus, 2012).

With the introduction of *Bt* cotton hybrids possessing resistance to American boll worm the area has been increasing and several reports are being added on the outbreak of various diseases on *Bt* cotton. The first *Bt* cotton hybrid was released by Monsanto-Mahyco Biotech during 2002. In Madhya Pradesh, the crop was afflected by the leaf curl virus (LCV), with 100 per cent infection. It is an irony that while some of the private hybrids and varieties released earlier were resistant to LCV, but *Bt* cotton was found to be susceptible to LCV. Simultaneously,

in the Vidarbha belt of Maharashtra, cotton crop planted over 30,000 ha has been widely affected because of the emergence of a disease of the roots called 'root rot'. It is believed that this disease is caused due to a mismatch of the *Bt* genes relevant in the USA and India. Many farmers have recorded only up to 50 per cent germination of seeds and many others had poor germination (Ranja Sengupta, 2002).

However, the production potential of the crop has not been fully exploited due to several biotic and abiotic factors. The crop suffers from many fungal diseases, of which foliar diseases take a heavy toll and among the diseases, *Alternaria* leaf spot causes yield losses up to 26 per cent (Chattannavar *et al.*, 2006). Even before the cultivation of *Bt* cotton, *Alternaria* leaf spot of cotton was one of the most important diseases noticed throughout the world.

There is a need to understand different aspects of *Alternaria* spp with respect to its genetic variability since not much work has been done on these aspects in the past. In addition, it helps in comprehensive understanding of causal the organism. Keeping this in view, the present investigations were under taken to study the molecular variability of *Alternaria* leaf spot of *Bt* cotton caused by *Alternaria* spp

MATERIALS AND METHODS

Molecular variability among the isolates of *Alternaria* spp on *Bt* cotton : Total genomic DNA from fungal isolates was extracted by using the CTAB DNA extraction protocol for plant DNA isolation from Saghai-Maroof *et al* (1984). The isolated DNA was then purified by Proteinase K and RNase A treatments for one hour. each, at 37 ^{0}C followed by one extraction with a 1:1 mixture of phenol: chloroform + isomyl alcohol (24:1 v/v) and two chloroform + isomyl alcohol (24:1 v/v) extractions. The DNA in the final aqueous layer was then precipitated by adding 1/10 times volume of 3M sodium acetate, pH 5.6 and two times with 70% ethanol, dried under vacuum and dissolved in minimum volume of 10:1 tris-EDTA, pH 8.0 buffer. The DNA concentration was estimated with a DNA fluorometer (Hoeffer Scientific, San Francisco, USA) using Hoechst 33285 as the DNA intercalating dye and calf thymus DNA as the standard (Brunk *et al.*, 1979). These estimates were confirmed by staining DNA with ethidium bromide after electrophoresis in 0.8 per cent agarose gel at 100 V for 1 hr. in Tris-acetate-EDTA (TAE) buffer (0.4 M Tris-acetate, 0.001 M EDTA, pH 8.0) using known DNA concentration standards (ë DNA, uncut).

PCR optimization and primer survey : The reaction condition for polymerase chain reaction (PCR) was optimized by varying concentrations of template DNA (10-60 ng), AmpliTaq DNA polymerase (0.5-2U) and Mg $^{++}$ salt (0-5 mm) in order to identify the most suitable RAPD primers for the study of molecular variations among the isolates. Thirteen random decamer primers from the OPA-02, OPA-03, OPA-07, OPC-05, OPC-06, OPC-15, OPD-07, OPM05, OPM-10, OPM-20, OPO-02, OPO-10 and OPO-16 (all primers from M/s Bangalore Genei, Pvt. Ltd. Bangalore) were surveyed and the primers generating highly polymorphic amplification products were identified and used for analyzing all the isolates.

PCR and agarose gel electrophoresis : PCR reactions were carried out in a DNA Thermal Cycler (Eppendorf, Master cycler gradient) each of the 25 µl reaction mixture contained 1X reaction buffer (10 mM Tris-Cl, pH 8.3 and 50

Random primers with following sequences were used in RAPD

Sl. No.	Primer	Sequence
1	OPA-02	TGCCGAGCTG
2	OPA-03	AGTCAGCCAC
3	OPA-07	GAAACGGGTG
4	OPC-05	GATGACCGCC
5	OPC-06	GAACGGACTC
6	OPC-15	GACGGATCAG
7	OPD-07	TTGGCACGGG
8	OPM-05	GGGAACGTGT
9	OPM-10	TCTGGCGCAC
10	OPM-20	AGGTCTTGGG
11	OPO-02	ACGTAGCGTC
12	OPO-10	TCAGAGCGCC
13	OPO-16	TCGGCGGTTC

mM KCl), 3 mM $MgCl_{2,}$ 1 U of Taq DNA polymerase; 200 µM each of dATP, dTTP, dCTP and dGTP (all reagents from M/S Bangalore Genei, Pvt. Ltd. Bangalore); 0.6 µM of primer and approximately 20 ng of template DNA. The PCR amplification conditions were as follows: Initial extended step of denaturation at 94 ^{0}C for 3 min followed by 40 cycles of denaturation at 94 ^{0}C for 1 min, primer annealing at 32 ^{0}C for 1 min and primer elongation at 72 ^{0}C for 1 min, followed by an extended elongation of step at 72^{0}C for 5 minutes. Reaction products were mixed with 2.5 µml of 10X loading dye (0.25% bromophenol blue, 0.25% Xylene Cyanol and 40% Sucrose, w/v) and spun briefly in a microfuge before loading on the gel (Sambrook *et al.*, 1989). The amplification products were electrophoresed on 1.2 per cent agarose gel at 100 volts. Gels were stained with ethidium bromide and photographed on poloroid 667 film under ultra violet light.

Scoring of amplified fragments : The amplified profiles for all the primers were compared with each other and bands of DNA fragment were scored as '1' for presence and '0' for absence, generating '0' and '1' matrix. Per cent polymorphism was calculated by using the formula.

$$\text{Per cent polymorphism} = \frac{\text{Number of polymorphic bands}}{\text{Total number of bands}} \times 100$$

Table 1. Banding profile of different primers for different isolates of *Alternaria* spp

Sl. No.	Primer	Total bands	Polymorphic bands	Per cent polymorphism
1	OPA-02	6	3	50.00
2	OPA-03	7	7	100.00
3	OPA-07	5	5	100.00
4	OPC-05	5	5	100.00
5	OPC-06	6	6	100.00
6	OPC-15	7	6	85.71
7	OPD-07	3	2	66.66
8	OPM-05	8	8	100.00
9	OPM-10	9	9	100.00
10	OPM-20	6	6	100.00
11	OPO-02	6	6	100.00
12	OPO-10	7	7	100.00
13	OPO-16	7	6	85.57

Analysis of the profile of the amplified fragments : Pair wise genetic similarities between isolates were estimated by DICE similarity coefficient. Clustering was done using the symmetric matrix of similarity coefficient and cluster obtained based on un weighted pair group arithmetic mean (UPGMA) using sequential agglomerative hierarchal nested (SAHN) cluster analysis of NTSYS-PC version 2.0 (Rohlf, 1998).

Internal Transcribed Spacer (ITS) – Polymerase Chain Reaction (PCR) : The ribosomal DNA (rDNA) unit contains genetic and non-genetic or spacer region. Each repeat unit consists of a copy of 18S, 5.8S and 28S like rDNA and its spacer like Internal Transcribed Spacer (ITS) and intergenic spacers (IGS). The rDNA

Table 2. Similarity co-efficient of seventeen isolates of Alternaria spp.

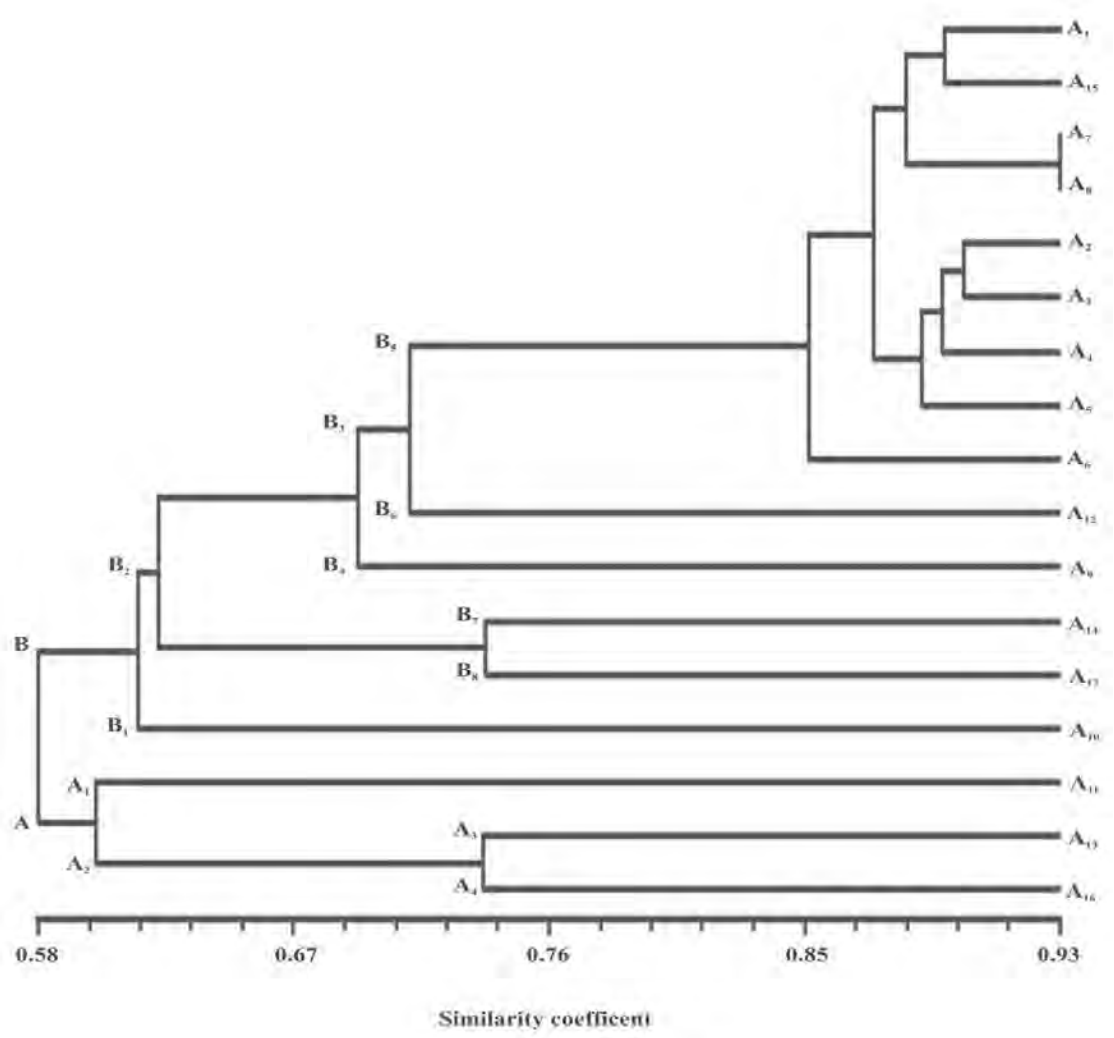
Isolates	A_1	A_2	A_3	A_4	A_5	A_6	A_7	A_8	A_9	A_{10}	A_{11}	A_{12}	A_{13}	A_{14}	A_{15}	A_{16}	A_{17}
A_1	1.00																
A_2	0.87	1.00															
A_3	0.90	0.90	1.00														
A_4	0.84	0.89	0.90	1.00													
A_5	0.87	0.89	0.88	0.89	1.00												
A_6	0.84	0.88	0.84	0.81	0.88	1.00											
A_7	0.89	0.89	0.88	0.87	0.87	0.88	1.00										
A_8	0.84	0.89	0.85	0.86	0.89	0.85	0.93	1.00									
A_9	0.71	0.75	0.71	0.70	0.71	0.64	0.71	0.73	1.00								
A_{10}	0.59	0.65	0.59	0.64	0.62	0.58	0.62	0.61	0.65	1.00							
A_{11}	0.63	0.65	0.68	0.62	0.58	0.59	0.63	0.59	0.44	0.46	1.00						
A_{12}	0.68	0.74	0.73	0.73	0.68	0.64	0.73	0.75	0.54	0.59	0.60	1.00					
A_{13}	0.59	0.51	0.56	0.56	0.55	0.55	0.59	0.60	0.40	0.44	0.56	0.58	1.00				
A_{14}	0.68	0.74	0.70	0.67	0.68	0.66	0.73	0.69	0.65	0.68	0.52	0.63	0.42	1.00			
A_{15}	0.89	0.85	0.86	0.85	0.87	0.80	0.89	0.89	0.69	0.63	0.66	0.69	0.59	0.68	1.00		
A_{16}	0.75	0.69	0.71	0.68	0.71	0.71	0.71	0.70	0.48	0.48	0.65	0.63	0.73	0.55	0.75	1.00	
A_{17}	0.57	0.62	0.59	0.55	0.57	0.55	0.57	0.58	0.50	0.53	0.45	0.53	0.41	0.73	0.55	0.42	1.00

have been employed to analyze evolutionary events because it is highly conserved, where as ITS rDNA is more variable. Hence, it has been used for investigation of these species level relationships.

ITS-PCR amplification of rDNA sequences for *Alternaria* species was conducted in 50 ml reaction volumes using conserved ITS1 and ITS4 primers (White *et al*, 1990). Each reaction consisted of 2 ml of 50 ng/ml DNA template, 5 ml of 10X PCR buffer, 0.5 ml of 25mM dNTPs, 1.5 ml of 15 mM MgCl2, 0.3 ml of 1.25U Taq DNA polymerase, 1 ml each of 10 mM primers ITS1 (5' TCC GTA GGT GAA CCT GCG G 3') and ITS 4 (5' TCC TCC GCT TAT TGA TAT GC 3 ') and 38.7ml sterile distilled water. The PCR protocol was standardized to amplify rDNA sequences from a strain each of *Alternaria* spp. infecting cultivated species of *Bt* cotton. The standardized protocol had cycling parameters of initial denaturation at 94°C for 4 minutes followed by 33 cycles of denaturation at 94°C for 1 min, annealing at 55°C for 1 min and extension at 72°C for 1.5 min. A final extension at 72°C for 5 minutes was done at the end of amplification. Negative controls were used to test for false priming and amplification. A 10-ml PCR amplification product for each of the *Alternaria* species was visualized in a 1.2 per cent agarose gel and viewed under UV light following staining with ethidium bromide.

RESULTS AND DISCUSSION

Molecular variability among the isolates of *Alternaria* spp on *Bt* cotton : The analysis of genetic variation in plant pathogen populations is an important prerequisite for understanding the evolution in the plant-patho system. Polymerase chain reaction (PCR) based molecular markers are useful tools for detecting genetic variation within populations of plant pathogens. Random Amplified Polymorphic DNA (RAPD) was used to detect the variation among the 17 isolates of *Alternaria* spp collected from different districts of North Karnataka. OPA, OPC, OPD, OPM and OPO series of primers obtained from Operon technologies, M/S Bangalore Genie,



A_1: UAS Dharwad
A_2: Annigeri
A_3: Dharwad Farm
A_4: Dharwad
A_5: Javalgeri
A_6: Kalmala
A_7: Gabbur
A_8: Sunkeshwarhal
A_9: Hattigudoor
A_{10}: Kannolli
A_{11}: Bijapur
A_{12}: Hanumanamatti
A_{13}:Yaragatti
A_{14} :Ulligeri
A_{15}: Hirebagevadi
A_{16}: Bagalkot
A_{17}:Dediapada (GJ)

Fig. 1a. Dendrogram based on RAPD analysis of 17 isolates of *Alternaria* spp.

were used to determine genetic diversity among the isolates to construct a dendrogram. The profile of amplicons of different primers for seventeen isolates of *Alternaria* spp. is given in Table 1 and Fig 1. Among the 13 primers used for amplification OPA-03, OPA-07, OPC-05, OPC-06, OPM-05, OPM-10, OPM-20, OPO-02 and OPO-10 showed cent per cent polymorphism. The isolates exhibited overall polymorphism of about 92.68%. Out of a total of 82 bands, 76 polymorphic bands were obtained. Out of 13 primers OPA-02, OPA-08, OPB-02 and OPF-06 showed 100 per cent polymorphism.

The banding profile per primer also varied

from minimum of 3 bands (OPD 07) to maximum of 9 bands (OPM 10). From the RAPD analysis, the results revealed that a total of 92.68 per cent polymorphism was found between the isolates, indicating there is a molecular variability among the isolates. Information on the banding pattern for all the primers was used to determine genetic distance between the isolates and to construct a dendrogram by using unweighted pair group arithmetic mean method (UPGMA). Based on the simple matching coefficient a genetic similarity matrix was constructed to access the genetic relativeness among the isolates. The genetic similarity co efficient of seventeen isolates based on RAPD analysis is given in Table 2 and Fig. 1a.

The similarity co-efficient ranged from 0.40 to 0.93. The maximum genetic similarity of 93 per cent was between Gabbur (A_7) and Sunkeshwarhal (A_8) isolates. There was 90 per cent similarity between Annigeri (A_2) and Dharwad Farm (A_3) isolates, where as least similarity (40 per cent) was observed between Hattigudoor (A_9) and Yaragatti (A_{13}) followed by least similarity (41%) was observed between Yaragatti (A_{13}) and Dediapada (A_{17}). Further, the dendrogram constructed by UPGMA from the pooled data clearly showed two major clusters *viz.*, A and B at a similarity co-efficient of 0.58 (Fig. 1). Cluster A was further sub grouped in to 2 sub clusters *viz.*, A1 encompassing Bijapur (A_{11}) isolate and A2 encompassing Yaragatti (A_{13}) and Bagalkot (A_{16}) isolates. Cluster B was further sub grouped in to 2 sub clusters *viz.*, B1 encompassing Kannolli (A_{11}) isolate and B2 encompassing remaining all the isolates Dediapada (A_{17}), Ulligeri (A_{14}), Hattigudoor (A_9), Hanumanamatti (A_{12}), Kalmala (A_6), Javalgeri (A_5), Dharwad (A_4), Dharwad Farm (A_3), Annigeri (A_2), Sunkeshwarhal (A_8), Gabbur (A_7), Hirebagevadi (A_{15}) and UAS Dharwad (A_1) isolates. In B2 sub cluster maximum similarity was found between Gabbur (A_7) and Sunkeshwarhal (A_8) isolates as well as Annigeri (A_2) and Dharwad Farm (A_3) isolates.

Polymerase chain reaction (PCR) based molecular markers are useful tools for detecting genetic variation within populations of plant pathogens. Random amplified polymorphic DNA (RAPD) markers have been widely used for estimating genetic diversity in natural populations (Annamalai *et al.*, 1995). The analysis of RAPD polymorphism in isolates of *Alternaria* spp from different agro climatic regions across North Karnataka revealed the occurrence of high level of polymorphism (92.68%) indicating wide and diverse genetic base.

Amplification of ITS1 and ITS4 region : The full length ITS rDNA region was amplified with ITS1 (5' TCC GTA GGT GAA CCT GCG G 3 ') and ITS 4 (5' TCC TCC GCT TAT TGA TAT GC 3 ') primers for among 17 isolates of *Alternaria* spp. Only 4 isolates were sent for confirmation at species level after the identification from Agharkar Research Institute, Pune. DNA amplicon was observed at the region 575 bp with concentration of around 150 ng/mg. The amplified products were checked on 1.2% agarose gel electrophoresis (Fig 2).

This seems to be the first report of amplification of ITS rDNA region of *Alternaria dianthi* isolated from *Bt* cotton in india. Since, there is less number of ITS rDNA region of isolate (A_2) *Alternaria dianthi* in gene bank itself.

Sequences of *Alternaria* spp ITS rDNA

DNA sequencing : The DNA sequences were obtained for ITS rDNA. The sequences of these isolates are given below.

$_{A2}$isolate,ITS-1

GAAAGGCGGGATGGACGGGCTGGATCT
CTCGGGGTTACAGCCTTGCTGAATTAT
TCACCCTTGTCTTTTGCGTACTTCTTG
TTTCCTTGGTGGGTTCGCCCACCACTA
GGACAAACATAAACCTTTTGTAATTGCA
ATCAGCGTCAGTAACAAATTAATAATTA
CAACTTTCAACAACGGATCTCTTGGTTC
TGGCATCAATGAAGAACGCCTTTGAATG
CGAAAGGGCTGTGAATTGCAGAATTCT
TTGAATCTTCAAATCTTTCAACGATTCT
TGCGCCCTTTGGTATTTCAAAGGGTTC
GTCTGTTCTGCCGTAACTTAGACACCC
GATGTTTGAATGTGGTATTATTAATTTG
TTACTGACTTTGATTGCAATTACAAAAA
GTTTATGTTTGTCCCAGTGGTGGGCGA
ACCCACCAAGGAAACAAGAAGTACCCC
CAAAGACAAAGGTGAATAAATCCACAAG
GGTGTTTTTTCCCAGAGATTCCCCCCC
CCCCTCATAGTTGTGTAATGAACCCTC
CCCAAGTTCACATACGGGAAAA

$_{A2}$isolate,ITS-4

GCCGGTAGGCGAGAGGCTGGCATCTCT
CGGGTGTACAGCGGCTTAATGGAAGCT
ACACCTTTGCTGAGCGAGAGTGCGACT
TGTGCTGCGCTCCGAAACCAGTAGGCC
GGCTGCCAATTACTTTAAGGCGAGTCT
CCAGCAAAGCTAGAGACAAGACGCCCA
ACACCAATCAAAGCTTGAGGGTACAAAT
GACGCTCGAACAGGCATGCCCTTTGGA
ATACCAAAGGGCGCAATGTGCGTTCAA
AGATTCGATGATTCACTGAATTCTGCAA
TTCACACTACTTATCGCATTTCGCTGC
GTTCTTCATCGATGCCAGAACCAAGAG
ATCCGTTGTTGAAAGTTGTAATTATTAA
TTTGTTACTGACGCTGATTGCAATTACA
AAAGGTTTATGTTTGTCCTAGTGGTGGG
CGAACCCACCAAGGAAACAAGAAGTAC
GCAAAAGACAAGGGTGAATAATTCCGC
AAGGCTGTTTCCCCCAGAGATTCCACC
CCGCCTTCATATTTGTGTAATGATCCC
TCCCCAAGTTCACCTACGAGAAAAA

$_{A5}$isolate,ITS's-1

AGCGGAAGGCGGGCTGGATCTCTCGGG
GTTACAGCCTTGCTGATTATTCACCCT
TGTCTTTTGCGTACTTCTTGTTTCCTTG
GTGGGTTCGCCCACCACTAGAGAAGCA
AAATACTTTTGTAATTGCAATCAGCGTC
CTAGAGAATTACAAATTACACCTTTCAA
CGCCTGATGGTATGGGGAGGGTTCGAT
GAAGAAGGCCTCGGAAAGGGAAAAGGC
GGCGAAGTGGGTTCTTTAAGGAAGGAT
TATTCATTGAAGCATTAACCGCCCTTTG
CGATTTCAGTTCCTTCGGTCATCAAAG
CGAGAACCAAAAAATCCATTCTGAAATT
GGTGTTGGTTAATTTGTCACTGACGCT
GATGGAAATTACCAAAGGTTTATTTGTG
TCCCGGTGGAGGGCGAACCGACCAACG
CAACATTAAAACCCCTAAAAACAAGGGT
AAAAAATCCACAAGGGCGGGAGCCGGA
AGAAGATCCCGCATCCGGTAAGATTAG
CGGCGGACCCCACGATAAGACACAGCG
GAGGAAAG

$_{A5}$isolate,ITS-4

ACGTAATGGGTAAAGTGAAAAATTAGGA
GGGCTCGGAGTCTCAGGTTTCAACCGC
TGCTTGGATGCTACACCTTTGCTGAGG
AGAGTGCGACTTGTGCTGCGCTCCGAA
ACCAGTAGGCCGGCTGCCAATTACTTT
AAGGCGAGTCTCCAGCAAAGCTAGAGA
CAATACACCCAACACCTTTCAAAGCTTG
AGGGTACAAATGACGCTCGAACAGGCA
TGCCCTTTGGAATACCAAAGGGCGGGT
GGTGCGTTAATCTTCGATGATTCAATGA
ATTCAACAAATCATACCACCTTTTGTAT
TTCCGTGCGTTTTTCCTTGAAGGCCGA
ACCCAGAAAATCCGTTGTTGAATTTTGT

ATTGATCATTTTGTAACGGACTTTGATG
GAAATTCCAAAAGGTTTATTTGGGCCCA
GGTGGTGGGGTAACCCACCCAATTTTT
TTTAACAATCCAAAAAAGCAAGGGTAAA
AAATCCATAAAGCTTTTTATCCAACTTT
ATATCCCCCCCCCCCACGTAAGTTTCCG
AAAGAACCTTACGCATATGTTTAAACGG
GAGAAAACG

$_{A8}$isolate,ITS’s-1

CAGCGGAAGCCGGGCTGGAATCTCTCG
GGGTTACAGCCTTGCTGAATTATTCAC
CCTTGTCTTTTGCGTACTTCTTGTTTCC
TTGGTGGGTTCGCCCACCACTAGGACA
AACATAAACCTTTTGTAATTGCAATCAG
CGTCAGTAACAAATTAATAATTACAACT
TTCAACAACGGATCTCTTGGTTCTGGC
ATCGATGAAGAACGCAGCGAAATGCGA
TAAGTAGTGTGAATTGCAGAATTCTGTG
AATCTTCGAATCTTTGAACGCACATTGC
GCCCTTTGGTATTCCAAAGGGCATGCC
TGTTCGAGCGTAATTTGTACCCTCAAG
CTTTGATTGGGGTTGGGCGTCTTGGCT
CTAGCTTTGCTGGAGACTTACCTTAAAG
TAATTGGCAGCCGGCCTACTGGTTTCG
GAGCGCAGGAAACAGTCGCACTCTCTA
AGAGCAAAGGTCTAGCATCCATTAAGC
CTTTTTTCAACTTTTGACCTCGCGCTC
CTTTATATATACCCGCTGAACCTACCC
ATATTTCTAATCCGAGAAAAAA

$_{A8}$isolate,ITS-4

ACAAGCGGGAATGGAGGGCTGGGTCTC
TCGGGTGTACAGCGGCTTAATGGAAGC
TACACCTTTGCTGAGCGAGAGTGCGAC
TTGTGCTGCGCTCCGAAACCAGTAGGC
CGGCTGCCAATTACTTTAAGGCGAGTC
TCCAGCAAAGCTAGAGACAAGACGCCC
AACACCTTTCAAAGCTTGAGGGTACAAA
TGACGCTCGAACAGGCATGCCCTTTGA
AATACCAAAGGGCGCAATGTGCGTTCA
AAGATTCGATGATTCACTGAATTCTGCA
ATTCACGCTACTTATCGCATTTCGCTG
CGTTCTTCAGCGATGCCAGAACCAAGA
CATCCGTTGTTGAAAGTTGTAATTATTA
ATTTGTTACTGACGCTGATTGCAATTAC
AAAAGGTTTATGTTTGTCCTAGTGGTGG
GCGAACCCACCAAGGAAACAAGAAGTA
CGCAAAAGACAAGGGTGAATAATTCAGC
AAGGCTGTTTCCCCGAGAGATTCCAGC
CCGCCTTCATATTTGTGTAATGATCCC
TCCGCAAGTTCACCTACGGAAAA

$_{A15}$isolate,ITS’s-1

ACGGGGAGCAGGGCTGGATCTCTCGGG
GTTACAGCCTTGCTGAATTATTCACCC
TTGTCTTTTGCGTACTTCTTGTTTCCTT
GGTGGGTTCGCCCACCACTAGGACAAA
CATAAACCTTTTGTAATTGCAATCAGCG
TCAGTAACAAATTAATAATTACAACTTT
CAACAACGGATCTCTTGGTTCTGGCAT
CGATGAAGAACGCAGCGAAATGCGATA
AGTAGTGTGAATTGCAGAATTCAGTGAA
TCATCGAATCTTTGAACGCACATTGCG
CCCTTTGGTATTCCAAAGGGCATGCCT
GTTCGAGCGTCATTTGTACCCTCAAGC
TTTGCTTGGTGTTGGGCGTCTTGTCTC
TAGCTTTGCTGGAGACTCGCCTTAAAG
TAATTGGCAGCCGGCCTACTGGTTTCG
GAGCGCAGCACAAGTCGCACTCTCTAT
CAACAAAGGTCTAGCATCCATTAAGGC
CTTTTTTCAACTTTTGACCTCGGATCAG
GTAGGGATACCCGCTGAACTTAACCAT
ATCAATAAGCGGAAAAAAAAA

$_{A15}$isolate,ITS-4

GTAGCCGGTCGGCAGAGCTGGAATCTC
TCGGGTGTACAGCGGCTTAATGGATGC
TAGACCTTTGCTGATAGAGAGTGCGAC
TTGTGCTGCGCTCCGAAACCAGTAGGC

CGGCTGCCAATTACTTTAAGGCGAGTC
TCCAGCAAAGCTAGAGACAAGACGCCC
AACACCAAGCAAAGCTTGAGGGTACAA
ATGACGCTCGAACAGGCATGCCCTTTG
GAATACCAAAGGGCGCAATGTGCGTTC
AAAGATTCGATGATTCACTGAATTCTGC
AATTCACACTACTTATCGCATTTCGCTG
CGTTCTTCATCGATGCCAGAACCAAGA
GATCCGTTGTTGAAAGTTGTAATTATTA
ATTTGTTACTGACGCTGATTGCAATTAC
AAAAGGTTTATGTTTGTCCTAGTGGTGG
GCGAACCCACCAAGGAAACAAGAAGTA
CGCAAAAGACAAGGGTGAATAATTCAGC
AAGGCTGTAACCCCGAGAGATTCCAGC
CCGCCTTCATATTTGTGTAATGATCCC
TCCCCAAGTTAACCTACGAAAAAAA

The DNA sequences of selected isolates were compared using bioinformatics tools like National Centre for Bioinformatics (NCBI) blast programme. Based on sequence comparison, the identification of *Alternaria* spp. isolates confirmed and all the ITS rDNA sequences of isolates were confirmed as one *Alternaria dianthi,* two *Alternaria* spp. and one *Alternaria alternata.* The list of isolates, Accession number, Maximum per cent homology and name identified are given in a Table 3.

These results are in conformity with the reports of Kadam (2005) who studied that the amplified products of ITS region of 11 fungal species from different crops, including strains of *R. solani, R. bataticola, A. macrospora* and *R. areola* reported in the present study, ranged between 569-575 bp, coinciding with the sizes obtained from similar fungal pathogens from other strains of the same species. Molecular techniques, if not alone, can be used in conjunction with classical methods where the latter approaches can at least narrow pathogen diagnosis to genus level. Once genus is narrowed by morphology, symptomatology, host-specificity, *etc.*, then PCR can be used to differentiate species (Chakrabarty *et al.* 2007).

Table 3. Comparison and identity of *Alternaria* spp of *Bt* cotton with referred gene bank

Isolates No.	ITS, Primers	Agarkhar Research Institute, Pune identified as	Gene Bank Accession number	Strains	Reference	Maximum (%) Homology with
A_2	ITS1	*Alternaria dianthi*	D38758	DA-1	Kusaba and Tsuge (1995)	95(%) with *A. dianthi*
	ITS4		AY154702	IA259	Ghosta (2002)	95(%) with *A. dianthi*
A_5	ITS1	*Alternaria* spp	FJ899921	MDF1.1	Shweta *et al.* (2009)	95(%) with *A. alternata*
	ITS4		AF397044	39/355	Konstantinova *et al.* (2009)	95(%) with *A. alternata*
A_8	ITS1	*Alternaria* spp	HM003680	SVJM015	Visalakchi *et al.* (2010)	95(%) with *A. alternata*
	ITS4		DQ156341	S	Chakrabarty *et al.* (2005)	95(%) with *A. alternata*
A_{15}	ITS1	*Alternaria alternata*	DQ156341	S	Chakrabarty *et al.* (2005)	95(%) with *A. alternata*
	ITS4		DQ156341	S	Chakrabarty *et al.* (2005)	95(%) with *A. alternata*

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Jasmonic acid and salicylic acid induced protection against cotton leaf curl disease

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ABSTRACT : The present study highlights the effectiveness of Jasmonic acid (JA) and Salicylic acid (SA) spray against CLCuD. Response of different American cotton accessions namely RS 921, LH 2076, PIL 8, Ankur 3028 BGII and a *desi* cotton variety LD 694 to JA and SA in the induction of proteins for protection against cotton leaf curl disease (CLCuD) was studied. At four to six leaf stage, potted plants of different cotton accessions were sprayed with different concentration of JA and SA *i.e* 50 µM, 100 µM, 150 µM and 200 µM. JA at 150 µM and SA at 200 µM caused maximum protein induction in all the treated cultivars *w.r.t* their control. JA was found to be more effective than SA in the induction of proteins. SDS-PAGE revealed induction of proteins in 15-45 kDa molecular weight range in treated samples as compared to their respective controls. JA @ 150 µM and SA @ 200 µM resulted in lower disease incidence as well as disease index. Disease incidence was recorded to be 37%, 30%, 30% and disease index was found to be 48%, 40%, 40% in RS 921, LH 2076, Ankur 3028 BGII at 150 µM concentration of JA whereas, at 200 ìM SA disease incidence of 48%, 36%, 34% and disease index of 57%, 50%, 50% was recorded in above mentioned accessions respectively. The respective controls exhibited higher values for disease incidence and disease severity. Latent carry over detection of symptomless plants treated with 150 µM of JA and 200 µM of SA through PCR amplification using DNA â specific primers confirmed the presence of virus in all the tested cotton accessions except LD 694 which depicted that induced proteins do not eliminate virus but might be playing a role in suppressing the proliferation of virus or might have kept virus in inactive state. JA and SA application thus, resulted in imparting tolerance with the induction of PR proteins but does not lead to complete resistance against the disease.

Key words: Cotton leaf curl disease, jasmonic acid, latent carry over, proteins, salicylic acid

Plants are constantly attacked by various types of pathogen as a result they have evolved a plethora of wide variety of defence mechanisms which can be either local, systemic, constitutive or inducible. One particular inducible systemic response, is Systemic Acquired Resistance (SAR). SAR refers to a distinct signal transduction pathway that plays an important role in the ability of plants to defend themselves against pathogens by the induction of various types of proteins and metabolites. Application of novel plant protection chemicals that act by stimulating the plant’s inherent disease resistance mechanisms by means of induction of various types of proteins thus could prove beneficial in disease management. It is found that certain natural and synthetic compounds e.g. Jasmonic acid (JA), Salicylic acid (SA) and their structural analogues are capable of activating the defense mechanisms in plant and prove helpful in conferring tolerance/resistance against various pathogens (Wang *et al.,* 2005). Vanwees (2000) in his work signified the importance of SAR inducers like salicylates and jasmonates against broad spectrum of pathogens. Moreover, chemical inducers of plant resistance

possess quite different mode of action as compared to fungicides. The latter products have direct toxic effect on pathogens; are noxious to environment; have narrow spectrum of defense; ensure short lasting protection (Kuc 2001). Thus, application of chemical inducers of resistance is an exciting new perspective to supplement the classical chemical means of disease control by providing both effective and ecologically-friendly plant protection. Present investigations were carried out to study the role of Jasmonic acid and Salicylic acid for the induction of resistance to cotton leaf curl disease CLCuD in cotton.

MATERIALS AND METHODS

Materials and treatment : Seeds of three Gossypium hirsutum accessions namely RS 921 (highly susceptible), LH 2076 (moderately resistant), PIL 8 (resistant) and one G. arboreum LD 694 (immune) possessing differential resistance to CLCuD used in this study were procured from Cotton Section, Department of Plant Breeding and Genetics, PAU, Ludhiana. A G. hirsutum hybrid Ankur 3028 BGII was provided by Ankur Seeds Pvt. Ltd. Twelve seeds of each cotton accession were sown in earthen pots in duplicate and kept in insect proof screen cages under natural conditions.

Treatment of seedlings with JA and SA : JA and SA @ 50 µM, 100 µM, 150 µM and 200 µM were applied as foliar spray with atomizer at 4-6 leaf stage of plants. Water sprayed plants served as controls.

Sample collection : For protein extraction, leaf samples were collected at 24, 48, 72, 96 hrs and a week interval of spray. Samples were brought to laboratory under refrigerated conditions and were stored at -80°C in deep freezer till further use.

Leaf tissue (0.2 g) was homogenized in 25 mM Tris HCl buffer (pH 8.0) in a pre-cooled pestle and mortar on ice. Homogenate was centrifuged at 10,000 rpm for 25 minutes at 40C. The supernatant was used as protein extract. Soluble proteins in supernatant were estimated by the method of Lowry et al., (1951) and expressed as mg/g fresh weight of tissues (mg/g fr. wt.).

Protein profiling using SDS-PAGE : Supernatant of different cotton accessions was subjected to SDS-PAGE (Walker 1996) for protein profiling. Protein samples were mixed with sample buffer (0.5 M Tris HCl, pH 6.8, 10 % SDS, Glycerol, 0.1% Bromophenol blue and 5% 2-Mercaptoethanol) in 1:1 ratio and kept in boiling water for 2 minutes and centrifuged. Supernatant (20 il) was taken and loaded in PAGE along with molecular marker (6-8kDa) and was run along with the samples. The Coomassie brilliant blue G 250 stained gel after destaining was preserved in 7 per cent acetic acid solution. The protein banding pattern of treated samples was analyzed.

Statistical analysis : Statistical analysis of the experimental data was done to test the significance of treatments using factorial completely randomized design. Critical differences were tested at 5 per cent level of significance.

Inoculation and disease assessment : A week after spraying with JA and SA, plants were exposed to viruliferous whiteflies. The colonies of viruliferous whiteflies, were reared and maintained on highly susceptible potted cotton plants in separate screen house. Six

whiteflies/plant were released for inoculation of CLCuV. CLCuD incidence and severity was calculated using the following formulae (Anonymous 2008):

$$\textbf{Disease Incidence (\%)} = \frac{Pi}{Pt} \times 100$$

Where,

Pi = Number of infected plants

Pt = Total number of plants

Disease index (%)

The plants were graded according to revised CLCuD scale described in AICCIP as given in Table 1(a) and (b)

$$\textbf{Disease index (\%)} = \frac{N1}{S1} \times \frac{S2}{N2} \times 100$$

Where,

N1= Number of plants in check

N2= Number of plants in test entry

S1= Sum of all infection grades in check

S2= Sum of all infection grades in test entry

Detection for the presence/absence of viral DNA in symptomless plants : Leaf samples from symptomless plants of different cotton accessions which were earlier treated with 150 ìM of JA and 200 ìM of SA and inoculated with viruliferous whiteflies were collected to detect the presence/absence of satellite DNA â through PCR amplification using DNA â specific primer pair. Total genomic DNA from symptomless cotton plants was isolated using the CTAB (Cetyl trimethyl ammonium bromide) method as reported by Shaghai-Maroof *et al.,* (1984). The primer pair used for cloning and sequencing of DNA â (Anup 2013) were used for *in vitro* PCR based amplification/identification of CLCuV DNA â (Table 2). Denaturation was done at 94°C and annealing at 56 °C for 1 minute; Elongation at 72 °C for 1minute (35 cycles) and final elongation at 72 °C for 7-10 minutes.

RESULTS AND DISCUSSION

Effect of JA and SA doses on total leaf protein content (mg/g fr. wt.) in different cotton accessions at various time intervals. : The data pertaining to changes in protein concentration recorded at periodical interval of 24 hrs till a week in response to various doses of JA and SA *i.e.* 50 µM, 100 µM, 150 µM and 200 µM revealed statistically significant differences amongst the various doses applied (Table 3, 4, 5, 6 and 7). Amongst the treatments applied, JA caused highest increase in protein content in accession LD 694 (2.3 fold) followed by PIL 8 (2 fold), RS 921 (2 fold), ANKUR 3028 BGII (1.6 fold), and LH 2076 (1.6 fold) whereas SA resulted in 1.4, 1.1, 1.2, 1.2 and 1.4 fold increase in protein concentration with respect to control in PIL 8, RS 921, ANKUR 3028 BGII, and LH 2076 indicating that JA is a better inducer of proteins.

Statistically significant differences in protein concentrations at different doses of each treatment were observed as indicated in Fig 1, 2, 3, 4 and 5 respectively JA resulted in mean maximum protein content *i.e* 15.1, 13.1, 15.0, 12.0 and 11.5 mg/g fr. wt. at 150 ìM whereas SA resulted in mean maximum protein value (9.0, 8.0, 8.6, 8.5 and 6.4 mg/g fr. wt.) at 200 ìM of SA when compared with control value of protein (5.8, 6.6, 5.1, 6.5 and 4.0 mg/g fr. wt.) in accession RS 921, LH 2076, PIL 8, ANKUR 3028 BGII and LD 694, respectively.

It was observed that 150 ìM of JA and 200 ìM of SA resulted in highest protein induction in all the treated accessions as compared to their controls. Comparative analysis of mean protein induction in different cotton accessions treated with 150 ìM of JA and 200 ìM SA after a week

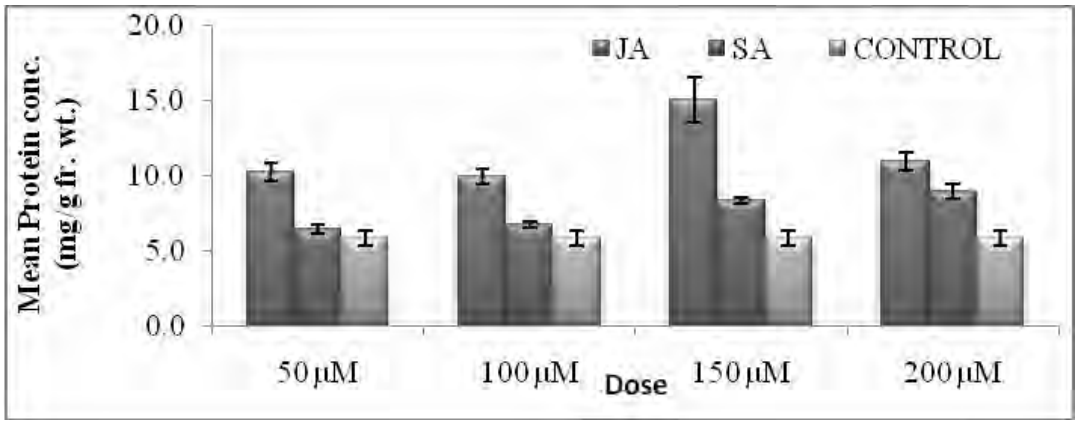

Fig 1 Effect of different doses of JA and SA on protein concentration of *G. hirsutum* accession RS 921
*Each value is a mean (Three replications) of values of protein concentration at different doses of each treatment

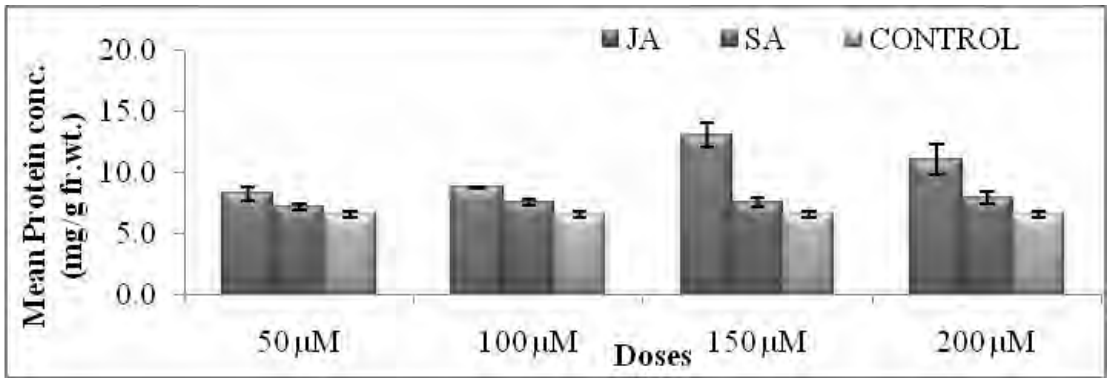

Fig 2 Effect of different doses of JA and SA on protein concentration of *G. hirsutum* accession LH 2076
*Each value is a mean (Three replications) of values of protein concentration at different doses of each treatment

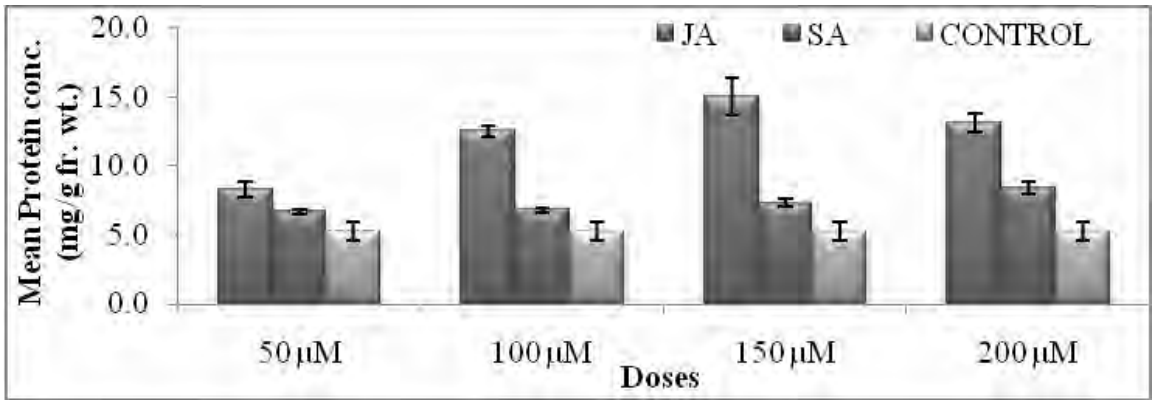

Fig 3 Effect of different doses of JA and SA on protein concentration of *G. hirsutum* accession PIL 8
*Each value is a mean (Three replications) of values of protein concentration at different doses of each treatment

interval as shown in Fig 6 revealed highest protein induction in RS 921 and PIL 8 followed by Ankur 3028 BGII and LH 2076. Similar results were obtained when cotton accessions were treated with 200 ìM of SA. Treatment with 150 ìM of JA resulted in 15.1, 15.0, 13.1, 12.0 and

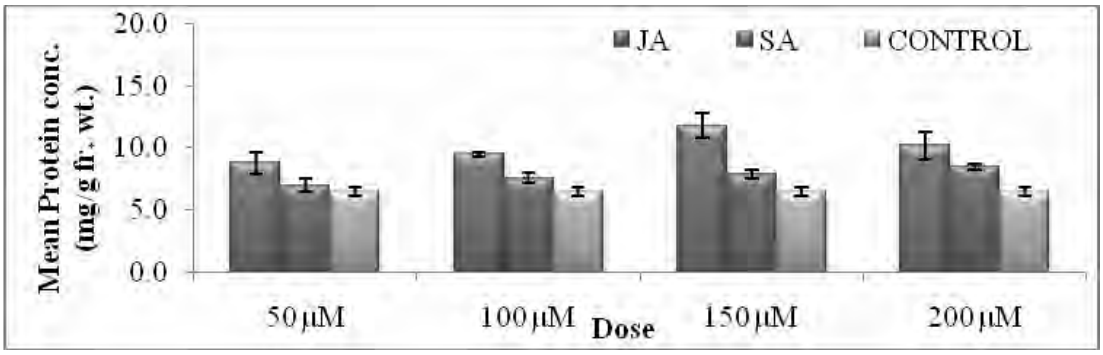

Fig 4 Effect of different doses of JA and SA on protein concentration of *G. hirsutum* accession Ankur 3028 BGII
*Each value is a mean (Three replications) of values of protein concentration at different doses of each treatment

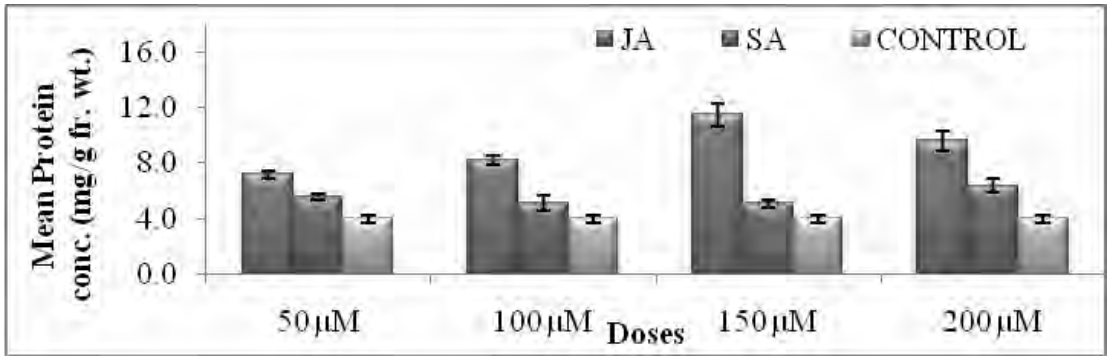

Fig 5 Effect of different doses of JA and SA on protein concentration of *G. arboreum* accession LD 694
*Each value is a mean (Three replications) of values of protein concentration at different doses of each treatment

11.5 mg/g fr. wt. protein in RS 921, PIL 8, LH 2076, Ankur 3028 BGII and LD 694 whereas 200iM of SA resulted in 9.0, 8.6, 8.0 , 8.5 and 6.4 mg/g fr. wt. protein in RS 921, PIL 8, LH 2076, Ankur 3028 BGII and LD 694 respectively as compared to protein values in control which were 5.8, 5.2, 6.6, 6.5 and 4.0 mg/g fr. wt.

This induction of proteins in different accessions is well supported with the fact that the known phytohormones JA and SA are found

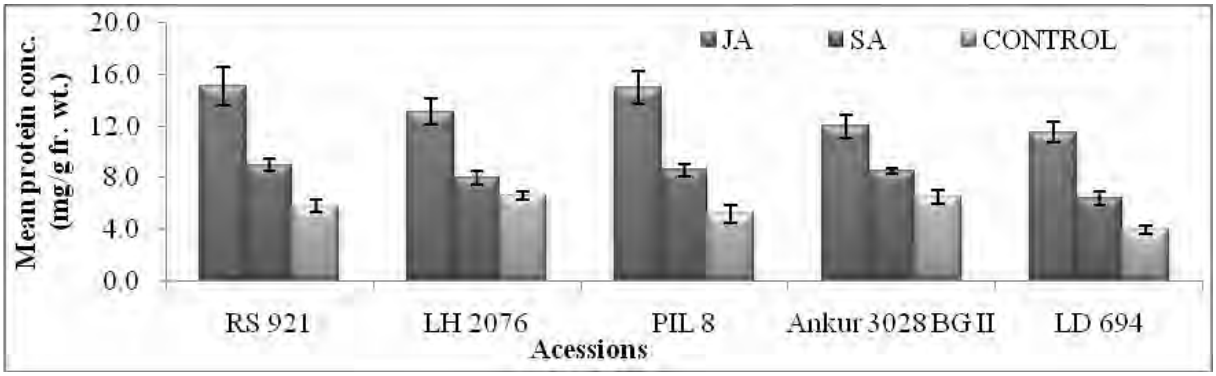

Fig 6 Comparative protein content in different cotton accessions with respect to 150 iM of JA and 200 iM of SA after a week interval

List of Abbreviations

AlMV	Alfalfa mosaic virus
BtMV	Beet mosaic virus
CLCuD	Cotton leaf curl disease
CLCuV	Cotton leaf curl virus
CMV	Cucumber mosaic virus
CTAB	Cetyl trimethyl ammonium bromide
DNA	Deoxyribonucleic acid
HCl	Hydrochloric acid
Hrs	Hours
JA	Jasmonic acid
JIP	Jasmonic acid induced proteins
kDa	Kilo Dalton
mg/g fr. Wt.	Milligram per gram of fresh weight
mM	Milimolar
PCR	Polymerase chain reaction
PR proteins	Pathogenesis-related proteins
Rpm	Revolutions per minute
SA	Salicylic acid
SAR	Systemic acquired resistance
SDS-PAGE	Sodium dodecyl sulphate polyacry lamide gel electrophoresis
TCV	Turnip crinkle viruse
ìM	Micromolar

Table 1 (a) Cotton leaf curl disease rating scale

Rating scale	Symptoms
0	Plants free from CLCuD
1	Thickening of small veins, only few upper leaves affected
2	Thickening of veins, curling and cupping of leaves
3	Thickening of veins, curling and cupping of leaves, enation development on underside of leaves
4	Thickening of veins, cupping, enations, stunting of plants and few bolls

Table 1 (b). Cotton leaf curl disease rating scale

Per cent disease index	Reaction
0	Highly resistant (HR)
0.01-5	Resistant (R)
5.1-25	Moderately Resistant (MR)
25.1-50	Moderately Susceptible (MS)
>50	Susceptible (S)

Table 2 Sequence DNA â specific primer pair used in the study

Source	Primer	Base sequence (5'-3')
AY083590.1	â 1 (F)	ACCGTGGGCGA GCGGTGCCCGAT
	â 1 (R)	CACGTGTAAT ACGTCTCCATCGTC

to regulate plant responses to biotic and abiotic stresses. JA and its metabolites are known to activate plant defence by the induction of jasmonate induced proteins and PR proteins (Delker 2006). Exogenous application of SA was shown to be mimicking certain aspects of a pathogen infection, resulting in SAR gene expression (Vernooij *et al* 1995). Uknes *et al* (1992) also stated the involvement of SA in SAR which activated gene expression of various defensive factors such as induction of PR proteins. Treatment with jasmonates was shown to induce various responses including the accumulation of serine proteinase inhibitors (Farmer 1990), leucine aminopeptidase and threonine deaminase (Hildmann *et al* 1992), phenylalanine ammonia-lyase, thionin (Anderson *et al* 1992), ribosome-inactivating protein (chaudhry *et al* 1994) and number of secondary metabolites were also shown to accumulate in cultured cells of various plant species upon treatment with JA (Gundlach 1992). It was found by Jing shi *et al.,* (2010) that exogenous application of JA and SA resulted in the induction of proteins in transgenic *Nicotiana benthamiana* with the expression of GhMPK7 gene. Haggag *et al.,* (2010) in their work reported that MeJA application on to the Beet mosaic virus (BtMV) infected sugarbeet plants resulted in the accumulation of total soluble proteins, chitinases etc which belong to various PR families. Yamada *et al.,* (2012) also reported that

treatment of rice plants with jasmonates results in the induction of OsJAZ8 protein. White (1979) reported the accumulation of PR proteins in tobacco plants which were treated with SA. SA and its structural analogue BTH are successfully used in inducing protection against various plant viruses like Tobacco mosaic virus (TMV) in tobacco, Turnip crinkle virus (TCV) in *Arabidopsis* (Lawton *et al* .,1996), and Cucumber mosaic virus (CMV) in tomato (Anfoka, 2000).

Comparative analysis of electrophoretic profile of cotton accessions treated with JA and SA : Extracted cotton leaf proteins were subjected to SDS-PAGE electrophoresis. Leaf samples of treated accessions namely RS 921, LH 2076, PIL 8, Ankur 3028 BGII and LD 694 which showed maximum protein induction at 150 µM of JA and 200 µM of SA were evaluated for protein profiling through SDS-PAGE. Total leaf proteins were resolved in molecular weights in the range 6-180 kDa *w.r.t* standard protein marker. Specific bands falling in the range of 6-49 kDa were reported in treated samples as compared to their respective control as shown in Plate 1 and Plate 2. It is known that PR proteins fall under the range of 15.8 kDa to 45 kDa (Van Loon and Antoniw 1982). Bol *et al* (1990) reported the induction of various PR proteins of molecular size ranging from 15.8-45 kDa with the application of SA in Tobacco plants. Jishan *et al* (2011) also revealed the induced expression of defence related genes like PR 2, PR 3, PR 5, PR 10 and Ta-JA2 which encode â,1-3 glucanase, chitinase, thaumatin-like protein, peroxidase etc. in different wheat accessions- namely Chinese Spring, Pumai 9 and Zhoumai 18 with MeJA treatment. Schweizer *et al* (1993) reported that in barley *(Hordeum vulgare* L.*)* application of Jasmonic acid effectively protected it against subsequent infection of *Erysiphe graminis* f.sp.

Table 3 Effect of different doses of JA and SA on leaf protein concentration (mg/g fr. wt. of *G. hirsutum* accession RS 921 recorded at periodic time intervals

Dose	Treatment	Time interval (h)					**Treatment mean**
		24	48	72	96	Week	
50 iM	JA	9.0	9.5	9.9	10.3	12.6	**10.3**
	SA	5.7	6.0	6.2	6.7	7.4	**6.4**
	Water	4.3	5.3	6.2	6.4	7.0	**5.8**
100 iM	JA	9.2	9.3	9.5	10.4	11.7	**10.0**
	SA	6.3	6.5	6.7	6.8	7.3	**6.7**
	Water	4.3	5.3	6.2	6.4	7.0	**5.8**
150 iM	JA	11.6	12.4	15.5	16.3	19.9	**15.1**
	SA	8.0	8.1	8.2	8.5	9.3	**8.4**
	Water	4.3	5.3	6.2	6.4	7.0	**5.8**
200 iM	JA	9.9	10.0	10.6	11.8	12.8	**11.0**
	SA	8.1	8.4	8.6	9.4	10.6	**9.0**
	Water	4.3	5.3	6.2	6.4	7.0	**5.8**

CD(0.05) Dose (A) = 0.056, Treatment (B) = 0.048, (A)(B) = 0.0097

Table 4 Effect of different doses of JA and SA on leaf protein concentration (mg/g fr. wt.) of *G. hirsutum* accession LH 2076 recorded at periodic time intervals

Dose	Treatment	Time interval (h)					**Treatment mean**
		24	48	72	96	Week	
50 iM	JA	7.2	7.5	8.1	8.5	10.3	**8.3**
	SA	6.6	6.8	7.1	7.5	7.9	**7.2**
	Water	6.0	6.3	6.5	6.6	7.6	**6.6**
100 iM	JA	8.5	8.7	8.8	8.9	9.0	**8.8**
	SA	6.7	7.2	7.6	8.0	8.3	**7.6**
	Water	6.0	6.3	6.5	6.6	7.6	**6.6**
150 iM	JA	11.0	11.9	12.7	13.3	16.7	**13.1**
	SA	6.6	7.0	7.5	8.0	8.7	**7.6**
	Water	6.0	6.3	6.5	6.6	7.6	**6.6**
200 iM	JA	8.8	9.0	9.8	13.3	14.7	**11.1**
	SA	6.8	7.1	7.9	8.9	9.3	**8.0**
	Water	6.0	6.3	6.5	6.6	7.6	**6.6**

CD(0.05) Dose (A) =0.039, Treatment (B) = 0.034, (A)(B) = 0.0.069

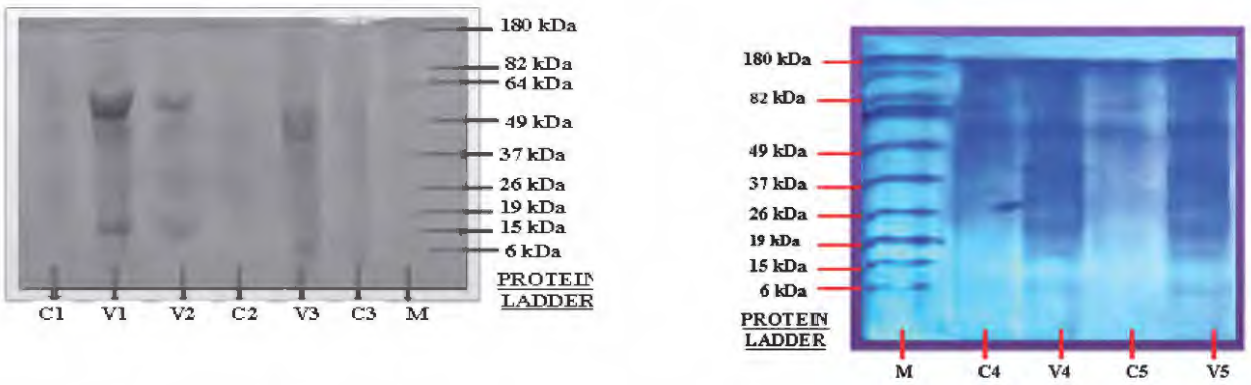

Fig 7. SDS-PAGE of leaf proteins of different cotton accessions at 150 iM of JA
M- Protein Ladder (6 kDa- 180 kDa), V1- Ankur 3028 BGII, V2- LH 2076, V3- LD 694, V4- RS 921, V5- PIL 8; C1, C2, C3, C4, C5 represent control of Ankur 3028 BGII, LH 2076, LD 694, RS 921 and PIL 8 accessions

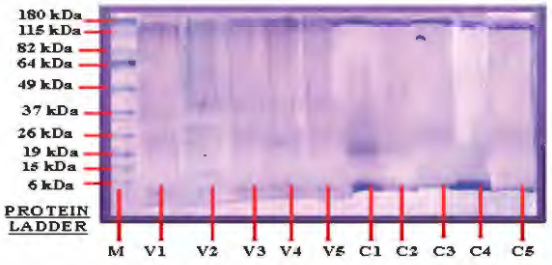

Fig 8. SDS-PAGE of leaf proteins of different cotton accessions at 200 iM of SA
M- Protein Ladder (6 kda- 180 kDa), V1- PIL 8, V2- RS 921, V3- LH 2076, V4- Ankur 3028 BGII, V5- LD 694; C1, C2, C3, C4, C5 – represent control of PIL 8, RS 921, LH 2076, Ankur 3028 BGII and LD 694 accessions

hordei by resulting in the formation of JA induced proteins (JIP) and Pathogenesis related proteins (PR). Two prominent groups of proteins of molecular size 25 kDa and 10 to 12 kDa sizes were induced. Exogenously applied MeJA imparted protection against *Alternaria brassicicola* in *Arabidopsis* by the induction of PDF1.2 PR-3 and PR-4 gene (Thomma *et al.*, 1998). Thus, it showed that exogenous application of JA and SA resulted in the induction of PR proteins of molecular size ranging from 15.8-45 kDa along with some other proteins as well in all the cotton accessions under treatment which might be including proteins specifically for virus control. The proteins acting against virus could include peroxidises, polyphenol oxidases or other PR proteins (Thaler *et al* 2002).

Effect of JA and SA on disease incidence and severity of CLCuD : Effect of JA and SA applied at a concentration of 150 µM on the incidence and severity of CLCuD. At 150 µM of JA and SA disease incidence and severity values were observed to be lower as compared to control in accessions RS 921, LH 2076 and Ankur 3028 BGII (Table 8). Disease incidence was (37%), (30%), (30%) and disease index was (48%), (40%), (40%) in RS 921, LH 2076, Ankur 3028 BGII at 150 µM concentration of JA which was (50%), (46%), (45%) and (78%), (65%), (63%) for disease incidence and disease index values in control. No disease was observed in PIL 8 and LD 694 cotton accessions.

Effect of 200 µM of JA and SA also

Table 5 Effect of different doses of JA and SA on protein concentration (mg/g fr. wt.) of *G. hirsutum* accession PIL 8 recorded at periodic time intervals.

Dose	Treatment	Time interval (h)					Treatment mean
		24	48	72	96	Week	
50 ìM	JA	6.9	7.6	8.2	8.7	9.9	**8.3**
	SA	6.3	6.4	6.7	6.7	7.3	**6.7**
	Water	3.3	4.3	5.8	6.2	7.0	**5.2**
100 ìM	JA	11.7	12.0	12.3	12.7	13.9	**12.5**
	SA	6.5	6.6	6.7	6.9	7.3	**6.7**
	Water	3.3	4.3	5.8	6.2	7.0	**5.2**
150 ìM	JA	11.9	13.3	14.1	16.1	19.6	**15.0**
	SA	6.7	6.9	7.2	7.7	8.2	**7.3**
	Water	3.3	4.3	5.8	6.2	7.0	**5.2**
200 ìM	JA	11.6	12.4	12.9	13.4	15.4	**13.1**
	SA	7.3	7.6	8.4	8.8	9.9	**8.6**
	Water	3.3	4.3	5.8	6.2	7.0	**5.2**

CD(0.05)
Dose (A) = 0.04, Treatment (B) = 0.03, (A)(B) = 0.07

Table 6 Effect of different doses of JA and SA on protein concentration (mg/g fr. wt.) of *G. hirsutum* accession Ankur 3028 BGII recorded at periodic intervals

Dose	Treatment	Time interval (h)					Treatment mean
		24	48	72	96	Week	
50 ìM	JA	7.1	7.9	8.0	9.0	12.0	**8.8**
	SA	6.0	6.2	6.5	7.7	8.6	**7.0**
	Water	5.9	6.0	6.3	7.0	7.5	**6.5**
100 ìM	JA	9.0	9.2	9.5	9.9	10.0	**9.5**
	SA	6.6	6.9	7.6	7.9	8.8	**7.6**
	Water	5.9	6.0	6.3	7.0	7.5	**6.5**
150 ìM	JA	9.6	10.8	11.1	12.6	15.0	**12.0**
	SA	7.0	7.5	7.9	8.3	9.0	**7.9**
	Water	5.9	6.0	6.3	7.0	7.5	**6.5**
200 ìM	JA	8.4	8.6	8.8	11.4	14.0	**10.2**
	SA	8.0	8.1	8.5	8.9	9.0	**8.5**
	Water	5.9	6.0	6.3	7.0	7.5	**6.5**

CD(0.05)
Dose (A) = 0.033, Treatment (B) = 0.029, (A)(B) = 0.058

indicated lower disease incidence and severity as compared to control (Table 9). At 200 ìM SA disease incidence was (48%), (36%), (34%) and

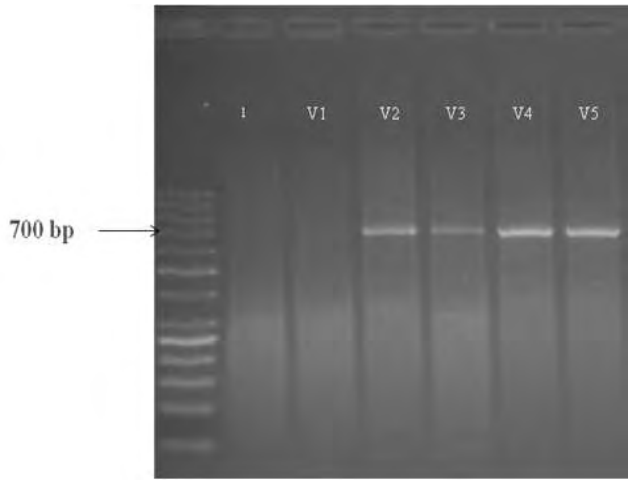

Fig 9 Detection of latent carryover of CLCuD in symptomless plants of different cotton accessions treated with 150 ìM of JA through PCR based amplification using primer â
1- Negative control, V1- LD 694, V2- RS 921, V3- LH 2076, V4- PIL 8, V5- Ankur 3028 BGII

Table 7 Effect of different doses of JA and SA on protein concentration (mg/g fr. wt.) of *G. arboreum* accession LD 694 recorded at periodic intervals

Dose	Treatment	Time interval (h)					Treatment mean
		24	48	72	96	Week	
50 ìM	JA	6.3	6.9	7.1	7.5	8.0	**7.2**
	SA	5.0	5.3	5.6	6.0	6.3	**5.6**
	Water	3.6	3.7	3.8	3.9	5.0	**4.0**
100 ìM	JA	7.2	7.7	8.3	8.8	9.0	**8.2**
	SA	3.9	4.3	4.6	5.6	6.9	**5.1**
	Water	3.6	3.7	3.8	3.9	5.0	**4.0**
150 ìM	JA	9.7	10.3	11.2	12.0	14.4	**11.5**
	SA	4.4	4.6	5.1	5.5	5.7	**5.1**
	Water	3.6	3.7	3.8	3.9	5.0	**4.0**
200 ìM	JA	7.3	8.8	10.0	10.4	11.5	**9.6**
	SA	5.4	5.7	6.0	6.6	8.1	**6.4**
	Water	3.6	3.7	3.8	3.9	5.0	**4.0**

CD(0.05)
Dose (A) = 0.041, Treatment (B) = 0.036, (A)(B) = 0.072

disease index was (57%), (50%), (50%) in RS 921, LH 2076, Ankur 3028 BGII which was (50%), (46%), (45%) and (78%), (65%), (63%) when

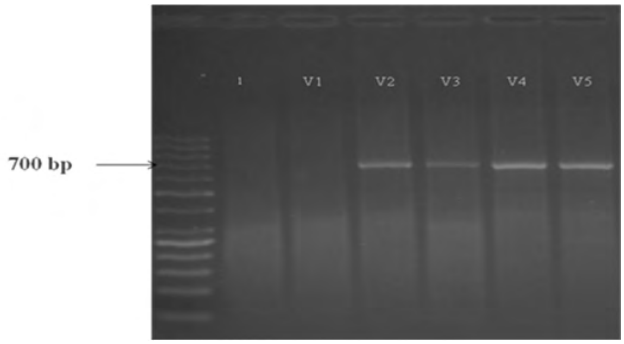

Fig 10 Detection of latent carryover of CLCuD in symptomless plants of different cotton accessions treated with 200 ìM of SA through PCR based amplification using primer â
1- Negative control, V1- LD 694, V2- RS 921, V3- LH 2076, V4- PIL 8, V5- Ankur 3028 BGII

compared with disease incidence and disease index values of control. Greater disease inhibition in disease incidence and severity was observed at 150 ìM of JA and 200 ìM of SA which was found to be negatively correlated with the amount of protein induced at these concentrations as at above mentioned concentration maximum protein induction was observed which could be responsible in lowering the disease. This decrease in disease parameters due to the induction of PR proteins and some other proteins is well supported by the findings of Chandra *et al* (2001) who showed that foliar spray of 0.02 per cent of SA twice at a week interval caused reduction in root rot disease caused by *Rhizoctonia solani* by enhancing the production of proteins. Spraying tobacco with SA induced the production of PR-1 proteins which played role in inhibiting the multiplication of alfalfa mosaic virus (AlMV) (Huijsduijnen *et al* 1986). Faheed *et al* (2006) reported that exogenous application of 0.1, 0.5 and 1mM SA

Table 8 Effect of JA and SA at a dose of 150 ìM on disease incidence and severity of CLCuD.

Cultivar	Disease Incidence (%) Dose @ 150 ìM									Disease Index (%)		
	14 *DAS			21 DAS			28 DAS					
	Control	JA	SA	Control	JA	SA	Control	JA	SA	Control	JA	SA
RS 921	20	0	37	40	27	50	50	37	75	78	48	60
LH 2076	0	0	0	20	25	37	46	30	62	65	40	53
PIL 8	0	0	0	0	0	0	0	0	0	0	0	0
Ankur 3028 BGII	0	0	0	28	20	33	45	30	55	63	40	50
LD 694	0	0	0	0	0	0	0	0	0	0	0	0

*DAS - Days after spray

Table 9 Effect of JA and SA at a dose of 200 ìM on disease incidence and severity of CLCuD

Cultivar	Disease Incidence (%) Dose @ 150 ìM									Disease Index (%)		
	14 *DAS			21 DAS			28 DAS					
	Control	JA	SA	Control	JA	SA	Control	JA	SA	Control	JA	SA
RS 921	20	0	12	40	35	32	50	45	48	78	50	57
LH 2076	0	0	0	20	16	15	46	33	36	65	48	50
PIL 8	0	0	0	0	0	0	0	0	0	0	0	0
Ankur 3028 BGII	0	0	0	28	25	28	45	32	34	63	45	50
LD 694	0	0	0	0	0	0	0	0	0	0	0	0

*DAS - Days after spray

on two week old plants of *Phaseolus vulgaris* induced partial inhibition in the accumulation of virus and elevated the induction of various total soluble proteins as compared to untreated plants which resulted in the induction of resistance against TNV. Resistance against the pathogen was attributed to the induction of proteins belonging to PR-2, PR-4 and PR-8 family.

Detection of latent carryover of CLCuD : Total DNA from symptomless plants of different cotton accessions which were earlier treated with 150 ìM of JA and 200 ìM of SA at which maximum protein induction was observed revealed the presence of a DNA â major band of about 600-700 bases in all the cotton accessions except LD 694 which indicated the presence of virus in all the cotton accessions except accessions *desi* cotton variety LD 694 (Fig 9 and Fig 10) which signified that PR proteins do not eliminate the virus. Earlier, Sabhiki *et al* (2004) also confirmed the presence of latent infection of CLCuV in apparently disease free plants and healthy plants of resistant genotypes.

The present study suggests that application of JA and SA results in the induction of proteins of different molecular weights which probably resulted in imparting tolerance against CLCuD whereas, PCR analysis showed that induced proteins were not able to eliminate the presence of virus particles. So, we can say that these chemicals can be used as an alternate to viricides in near future.

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Detection of pink bollworm, *Pectinophora gossypiella*, Saunders infestation using Soft X ray machine

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ABSTRACT : An experiment was carried on standardization of X ray radiography methodology for the detection of pink bollworm infestation in cotton bolls during 2012-2014 at Indian Institute of Crop Processing Technology, Thanjavur, Tamil Nadu, India. Studies revealed that the controllable input electrical parameters of the X ray machine *viz.*, voltage, current and exposure period required for the detection of internal infestation varied widely for cotton bolls compared to stored grains and fruits tested by other scientists. High voltage and current were required for dense cotton bolls to ensure adequate penetration of radiation. It was observed on visual analysis that the X ray radiation generated at 80 kV and 10 mA for 30 seconds resulted in the best visual images to view internal content of cotton bolls and observed to be the best for cotton bolls imagery out of 96 combinations tested for best detection of hidden infestation. While other combinations, for example, 60Kv, 4mA for 10 seconds and 90 Kv, 10mA for 30 seconds manifested into lighter and darker images, respectively.

Key words : Infestation, non destructing sampling, pink bollworm, X ray

Cotton, *Gossypium hirsutum* L., popularly called as "King of fibres" or "White gold" and it is cultivated in an area of 116.14 lakh hectare with an average production of 334 lakh bales in India (Anonymous, 2013). About 130 different species of insects and mites are reported to cause damage to cotton crop in India (Agarwal *et al.,* 1984). Among these, the bollworms *viz.,* american bollworm, *Helicoverpa armigera* (Hubner), spiny bollworm, *Earias insulana* (Boisdual), spotted bollworm, *Earias vitella* (Fabricius), pink bollworm, *Pectinophora gossypiella* (Saunders) pose greater threat to cotton production. Among the bollworms, the pink bollworm assumed major pest status in recent past (Gutierrez *et al.*, 2006). Further pink bollworm has become economically the most destructive and quarantine insect pest of cotton. Infestation of pink bollworm in cotton bolls cannot be seen through naked eyes because, the nature of PBW was soon after hatching larva enters the developing bolls through tip portion and entrance hole is closed as the boll mature. Therefore, infestation could not be seen, a kind of hidden infestation. Destructing sampling was the only conventional method for assessment of cotton boll damage. Hence, considering the importance and usefulness of non-destructive method of detecting (Milner *et al.*,1950; Schatzki and Fine, 1988; Haff and Slaughter, 1999) a laboratory experiment was undergone using X ray radiation.

X ray radiography provides a permanent, visible film record of the internal condition of the boll sample. It is especially useful for the rapid examination of relatively large samples to determine the extent of insect damage. X rays pass readily through the objects, although some of the radiation is absorbed and the amount of absorption depends on the density of the

material, its thickness as well as the voltage applied to generate the X rays. For example, more radiation will pass through the areas containing hollow portions caused by insect tunneling than through the surrounding areas since the insect tunneling reduces the total thickness of the exposed material. High voltages are required to generate adequate penetration through very dense material. Adjustment of the voltage, current and exposure period to the specific material is important because it affects the contrast of the image recorded on X ray sensitive film after exposure. If the voltage, current and exposure period is too high, too much of the radiation will pass through the exposed material and obscure differences in thickness within the material. Similarly, contrast will be poor if the voltage, current and exposure period is too low since too little radiation will pass through the material to form a usable image (Ramakrishnan *et al.*, 2011). Although th technology is known and the suitable X ray machinery available, the input factors *viz.*, the voltage, current and the exposure period have not been standardized for cotton bolls. In the absence of standardized values of these input factors, users resort to standardization every time. Standardizing the methodology for a cotton boll is quite an important task to further the use of X ray radiography. Therefore, considering the importance and usefulness of this non-destructive method of detecting insect damage levels of pink bollworm in cotton bolls using soft X ray machine.

MATERIALS AND METHODS

Laboratory experiment for standardization of X ray radiography were proposed to be undertaken on the pink bollworm infestation in cotton bolls was conducted at Indian Institute of Crop Processing Technology, Ministry of Food Processing Industries, Thanjavur, Tamil Nadu, India. Cotton bolls for experimentation were collected from experimental farm, Tamil Nadu Agricultural University, Coimbatore.

Description and functioning of equipment X ray high tension transformer : The High Tension (HT) Transformer generated the high voltages required for the X ray generation. Transformer generated can be upto 160kV. The transformer was fully immersed in special purpose high electrical insulation transformer oil. The transformer was housed inside a separate chamber on wheels for easy movement. The output of the HT transformer was taken to the X ray tube head in the inspection Chamber using custom made insulated cables.

X Ray tube head : Tube head was internally cooled using HT oil. The tube head was coated with additional lead lining to prevent X ray leakage. The Tube Head was from where the X rays are generated.

Sample inspection chamber : The inspection chamber were samples are kept for inspection one after the other. This chamber was fully lead lined inorder to prevent X rays from escaping into the operator area. The X ray chamber has the X ray tube head, from where X rays emanate. It has the object plate, with sensor area marked. The door to the chamber was also lead lined. The door also has a leaded glass window, which was usefull to view the exact position for placing the object in the chamber. An interlocking limit switch was provided, which ensures that the X rays can be switched ON only when the window was closed. During exposure,

window should be closed. The X ray protection for the X ray chamber was as per International Standard GB 18871.

X ray sensor : The X ray sensor was housed below the inspection chamber. Sensor was a digital sensor with 5 Mega Pixel resolutions. The area covered by the sensor was 150 x 150mm and was marked on the object plate. The output of the X ray sensor was provided to the PC using an USB port. The sensor was supported by software, where the parameters are changed for effectively viewing cotton bolls. The present investigations were intended to establish a methodology to facilitate the standardization of the X ray radiography and the input electrical parameters *viz.,* voltage (kV), current (mA) and exposure period (s). Required adjustments in voltage (kV), current (mA) and exposure period (s) in the X ray machine were effected during the exposure procedure. Once the exposure to radiation was done, the X ray film was processed with the help of chemicals for image development and image fixing.

Treatments and experiment procedure : Scientists at IICPT laboratories have found that fruit and vegetables require soft X ray radiations ranging from 60 to 90 kV at a current of 1 to 10 mA. The period of exposure also ranges between 10 and 55 seconds. Therefore, experiments were planned for cotton boll to find out a standard voltage, current and exposure period. The combination of 96 treatments was worked out and the same combinations were given. Image analysis was taken up for different combinations to find out and standardize the right one that gives us internal view of the cotton boll. Adjustments in combinations of current and exposure periods were made based on the preliminary image results while working in the laboratory.

RESULTS AND DISCUSSION

Out of ninety six different combinations of voltage, current and exposure period tried for cotton bolls infested with pink bollworm it was observed on visual analysis that the X ray radiation generated at 80 kV and 10 mA for 30 seconds resulted i the best visual images to view internal content of cotton bolls and observed to be the best for cotton bolls imagery out of 96 combinations tested for best detection of hidden infestation (Table 1). While other combinations, for example, 60Kv, 4mA for 10 seconds and 90 Kv, 10mA for 30 seconds manifested into lighter and darker images, respectively (Plate.1). In the plate 1, image with the combinations of 80 kV and 12 mA for 30 seconds, shows that the boll (bottom - left side) was found to be infested with 2^{nd} instar PBW larva. Presence of insect stage will hide the internal content of the boll (enlarged image).

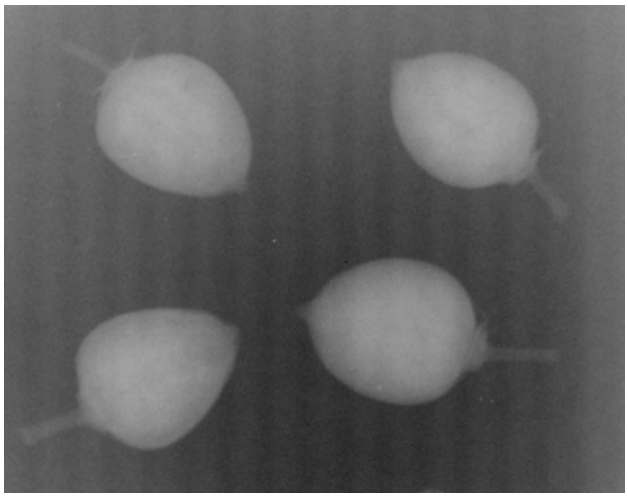
X ray radiography method is also used in detecting the hidden insect infestatio for it is also a non-destructive method. These techniques of X ray radiography were effectively used by Sarath Babu (1997) during quarantine processing of the germplasm imported from different countries and detection of several bruchids and chalcid species which were not reported from India. Similarly, this methodology was also used by Thomas *et al.,* (1995) on mango fruit infested with nut weevil, Shahin *et al.,* (2002) on apple, Karunakaran *et al.,* (2003a and 2003b) on western red spring wheat, on cherries and apple and on various fruits. Further investigations are necessary to carry out the brief usage of X ray radiography in detection of internal infestation of pink bollworm in cotton bolls. This study holds the basic research on non

Table 1. Treatment combinations of voltage, current and exposure period

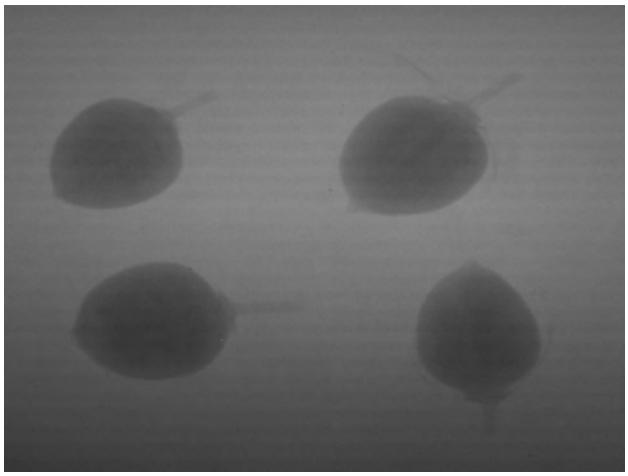
S. no	Kv	mA	S	S. no	Kv	mA	S	S. no	Kv	mA	S	S. no	Kv	mA	S
1	60	4	10	**26**	70	4	10	**51**	80	4	10	**76**	90	4	10
2	60	4	15	**27**	70	4	15	**52**	80	4	15	**77**	90	4	15
3	60	4	20	**28**	70	4	20	**53**	80	4	20	**78**	90	4	20
4	60	4	25	**29**	70	4	25	**54**	80	4	25	**79**	90	4	25
5	60	4	30	**30**	70	4	30	**55**	80	4	30	**80**	90	4	30
6	60	6	10	**31**	70	6	10	**56**	80	6	10	**81**	90	6	10
7	60	6	15	**32**	70	6	15	**57**	80	6	15	**82**	90	6	15
8	60	6	20	**33**	70	6	20	**58**	80	6	20	**83**	90	6	20
9	60	6	25	**34**	70	6	25	**59**	80	6	25	**84**	90	6	25
10	60	6	30	**35**	70	6	30	**60**	80	6	30	**85**	90	6	30
11	60	8	10	**36**	70	8	10	**61**	80	8	10	**86**	90	8	10
12	60	8	15	**37**	70	8	15	**62**	80	8	15	**87**	90	8	15
13	60	8	20	**38**	70	8	20	**63**	80	8	20	**88**	90	8	20
14	60	8	25	**39**	70	8	25	**64**	80	8	25	**89**	90	8	25
15	60	8	30	**40**	70	8	30	**65**	80	8	30	**90**	90	8	30
16	60	10	10	**41**	70	10	10	**66**	80	10	10	**91**	90	10	10
17	60	10	15	**42**	70	10	15	**67**	80	10	15	**92**	90	10	15
18	60	10	20	**43**	70	10	20	**68**	80	10	20	**93**	90	10	20
19	60	10	25	**44**	70	10	25	**69**	80	10	25	**94**	90	10	25
20	60	10	30	**45**	70	10	30	**70**	80	10	30	**95**	90	10	30
21	60	12	10	**46**	70	12	10	**71**	80						
22	60	12	15	**47**	70	12	15	**72**	80						
23	60	12	20	**48**	70	12	20	**73**	80						
24	60	12	25	**49**	70	12	25	**74**	80						
25	60	12	30	**50**	70	12	30	**75**	80						

destructing sampling method or X ray radiography. However there was a change in current and exposure period which may be attributed to built-in minor variations in different X ray machine used in experimentations. In future, we can expect precise work on detection of internal infestation of pink bollworm. Extensive work has been reported on the use of X rays to detect infestations in stored products due to internal grain feeders, the granary weevil, *Sitophilus granarius* Linnaeus, the rice weevil, *Sitophilus oryzae* L., the maize weevil, *Sitophilus zeamais* Mots., and the Angoumois grain moth, *Sitotroga cerealella* (Olivier) ingrain kernels by visual examination of the X ray radiographs (Keagy and Schatzki, 1991; Fenton and Waite, 1932). Only a few studies have used image processing algorithms to identify the insect infested wheat kernels using digital images of kernels (Karunakaran *et al.*, 2000; Keagy and Schatzki, 1993). From this context, the present findings are that X ray generated at 80 kV and 10 mA for 30 seconds resulted in the best visual images to view internal content of cotton bolls. Whereas, quarantine workers in India traditionally used only a range of 10 KV to 30 KV and a current of 4mA to 12mA for an exposure period of 10-25 seconds.

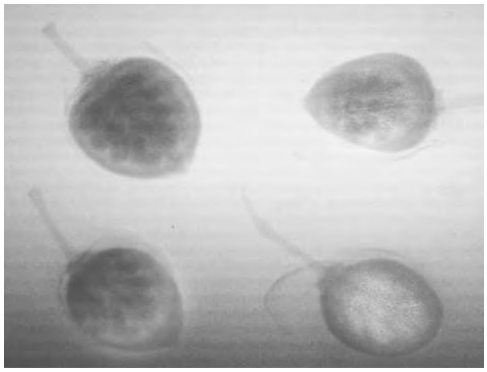
The range of X ray radiography values of about 20-34 KV, 12-30 mA current as optimum for the detection of stone weevil infestation in mango fruit. The present finding will form basis for Cotton Entomologist who want to work on



A) 1). 60 Kv 2). 4 mA 3). 10 s



B) 1). 90 Kv 2). 12mA 3). 30 s



C) 1). 80Kv 2). 12mA 3). 30 s

Plate 1. X ray radiography images of cotton with different combinations of A, B and C

detection of hidden infestation of cotton pink bollworm using soft X ray.

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Emerging insect pests of *Bt* cotton in middle Gujarat

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To keep pace with increasing global need of cotton and food security, researchers have devised various technologies to make the cotton production more economic and sustainable. Efforts are being made to validate and disseminate these technologies among farmers. While extending these technologies there is a need to keep vigilance on the emerging pests under the changing cropping system and issue the periodical advisories for timely IPM intervention.

Due to changes in the agroecosystem, high inputs, reduced numbers and volume of insecticidal sprays, number of new insect pests are now claiming *Bt* cotton as their new host. In many parts of the country and the world, *Bt* cotton is under threat due to unusual attack of some insect pests. There are the possibilities of spread of the pests which may make a havoc, if due attention is not being given. In middle Gujarat,

Table 1. Activity of emerging insect pests in *Bt* cotton fields located in Vadodara and Kheda district during 2014 -2015 (Averaged over 18 fixed plots)

Month and week		Number of Mirid bug/ plant		Infested fruiting bodies due to pink bollworm (%)			
				Square		Green boll	
		Vadodara district	Kheda district	Vadodara district	Kheda district	Vadodara district	Kheda district
1		2	3	4	5	6	7
November	**II**	0.00	0.81	2.36	1.66	2.50	1.80
	III	0.00	0.00	2.51	0.09	2.98	0.05
	IV	0.00	0.08	2.93	0.16	3.50	0.12
December	**I**	0.00	0.28	2.87	1.18	4.25	0.96
	II	0.00	0.23	3.46	0.72	6.78	0.48
	III	0.00	0.18	4.23	0.34	12.33	0.28
	IV	0.00	0.13	5.46	0.31	17.69	0.19
January	**I**	0.00	0.00	6.77	0.84	20.21	0.23
	II	0.00	0.11	9.69	3.11	26.43	1.63
	III	0.00	0.02	14.6	8.84	30.15	1.90
	IV	0.00	0.03	15.3	3.25	32.28	0.88
	V	0.00	0.00	17.7	1.52	36.95	0.24
February	**I**	0.00	0.00	0.04	0.00	41.03	0.28
	II	0.00	0.00	0.00	0.00	42.67	0.37
	III	0.00	0.00	0.00	0.00	47.79	0.54
Average	**0.00**	**0.12**	**5.86**	**1.47**	**21.84**	**0.66**	
SD	0.00	0.21	5.81	2.30	16.07	0.63	

Note: SD: Standard deviation

Table 2. Moth catches of pink bollworm in pheromone traps installed in *Bt* cotton fields located in Vadodara and Kheda district during 2014 - 15 (Averaged over 9 fixed plots)

Month and week		Average number of pink bollworm moth catches/ trap/ week	
		Vadodara district	Kheda district
1		2	3
November	**III**	0.11	0.22
	IV	0.00	0.17
December	**I**	0.00	0.06
	II	0.78	1.44
	III	2.94	1.89
	IV	4.33	0.61
January	**I**	3.28	2.00
	II	1.33	1.33
	III	2.39	1.67
	IV	2.11	2.33
	V	2.39	1.28
February	**I**	2.00	2.00
	II	2.72	0.89
	III	1.61	1.61
Average	**1.86**	**1.25**	
SD	1.31	0.75	

Note: SD: Standard deviation

the mirid bug and pink bollworm are found as emerging inset pests of *Bt* cotton during 2014-2015.

The cotton mirid bug, *Creontiades biseratense* (Distant) is an emerging insect pest on *Bt* cotton in Karnataka, India causing heavy shedding of squares and bolls which lead to significant reduction in seed cotton yield (Patil *et al.*, 2006; Ravi, 2007 and Udikeri *et al.*, 2009). The pest has also been noticed in Tamil Nadu, Andhra Pradesh and Maharashtra (Surulivelu and Dhara jothi, 2007).

Crops genetically engineered to produce insecticidal proteins from the bacterium *Bacillus thuringiensis* (*Bt*) kill some key pests and reduce reliance on insecticide sprays. Such *Bt* crops were commercialized in 1996 and planted on more than 50 million hectares worldwide in 2009 (James, 2009). The major threat to the continued success of *Bt* crops is evolution of resistance by pests. While most target pest populations remain susceptible, resistance to *Bt* crops has been reported in one of the most devastating pests of cotton globally recently, the pink bollworm (*Pectinophora gossypiella* Saunders), evolved resistance to transgenic cotton that produces *Bt* toxin Cry1Ac in western India (Bagla, 2010). Thus, the pink bollworm had the genetic potential to evolve resistance and was under intense selection for resistance in the field. The pink bollworm said to be rampant through out Vadodara district especially in middle Gujarat.

Department of Agriculture and Co operation, Ministry of Agriculture, Government of India sponsored project "On-line Pest Monitoring and Advisory Services under commercial crops (OPMAS)" on ICT based e-pest surveillance and advisory and implemented by NCIPM, New Delhi in 2014-2015. The main object is to monitor the pests in cotton, issue the pest advisories timely to extension agencies and farmers, disseminate the IPM activities across the country.

For the purpose, the investigation was undertaken during cropping period in different *Bt* cotton growing districts *viz.*, Vadodara and Kheda. In each district, major cotton growing three talukas were selected and in each taluka, three villages were selected as per the guideline. During the course of study total 18 villages of both the districts were surveyed at weekly interval by field assistants allotted to each taluka for recording the insect pests observations and recorded pest data were uploaded on the NCIPM website. In the present investigation, pink bollworm and mirid bug were found as the emerging pests in *Bt* cotton. The observations

Table 3. Infestation of pink bollworm, *P. gossypiella* in *Bt* cotton in Vadodara district during 2014 - 2015 (Boll Destruction Method)

Taluka	Village	Field no.	No. of damaged bolls out of 30			Picking wise infestation of pink bollworm (%)			
			1st	2nd	3rd	1st	2nd	3rd	**Average**
Karjan	Kandari	F1	03	25	12	10.00	83.33	73.33	**55.55**
		F2	04	19	28	13.33	63.33	93.33	**56.66**
		R1	04	26	19	13.33	86.66	63.33	**54.44**
		R2	07	25	25	23.33	83.33	83.33	**63.33**
					Average	**15.00**	**79.16**	**78.33**	**57.50**
	Bamangam	F1	00	16	12	0	53.33	40.00	**31.11**
		F2	01	05	15	3.33	16.66	50.00	**23.33**
		R1	02	12	10	6.66	40.00	33.33	**26.66**
		R2	02	04	23	6.66	13.33	76.66	**32.22**
					Average	**4.16**	**30.83**	**50.00**	**28.33**
	Ganapatpura	F1	00	25	10	0	83.33	33.33	**38.89**
		F2	00	24	27	0	80.00	90.00	**56.67**
		R1	03	20	25	10.00	66.66	83.33	**53.33**
		R2	00	22	08	0	73.33	26.66	**33.33**
Dabhoi					**Average**	**2.50**	**75.83**	**58.33**	**45.55**
	Kuvarpura	F1	05	08	10	16.66	26.66	33.33	**25.55**
		F2	08	14	07	26.66	46.66	23.33	**32.22**
		R1	00	05	12	0	16.66	40.00	**18.89**
		R2	00	08	10	0	26.66	33.33	**20.00**
					Average	**10.83**	**29.16**	**32.50**	**24.16**
	Khanpura	F1	04	03	15	13.33	10.00	50.00	**24.44**
		F2	00	09	10	0	30.00	33.33	**21.11**
		R1	00	12	17	0	40.00	56.66	**32.22**
		R2	3	04	10	10.00	13.33	33.33	**18.89**
					Average	**5.83**	**23.33**	**43.33**	**24.17**
	Menpura	F1	00	06	05	0	20.00	16.66	**12.22**
		F2	2	04	15	6.66	13.33	50.00	**23.33**
		R1	00	03	15	0	10.00	50.00	**20.00**
		R2	00	10	10	0	33.33	33.33	**22.22**
					Average	**1.67**	**19.17**	**37.50**	**19.44**
Savli	Gulabpura	F1	00	05	05	0	16.66	16.66	**11.11**
		F2	00	07	09	0	23.33	30.00	**17.78**
		R1	00	01	10	0	3.33	33.33	**12.22**
		R2	02	06	06	6.66	20.00	20.00	**15.55**
					Average	**1.67**	**15.83**	**25.00**	**14.16**
	Ghothada	F1	00	07	09	0	23.33	30.00	**17.78**
		F2	02	06	04	6.66	20.00	13.33	**13.33**
		R1	00	03	08	0	10.00	26.66	**12.22**
		R2	00	01	04	0	3.33	13.33	**5.55**
					Average	**1.67**	**14.17**	**20.83**	**12.22**
	Anjesar	F1	00	03	03	0	10.00	10.00	**6.67**
		F2	01	02	10	3.33	6.66	33.33	**14.44**
		R1	01	03	07	3.33	10.00	23.33	**12.22**
		R2	00	04	06	0	13.33	20.00	**11.11**
					Average	**1.67**	**10.00**	**21.67**	**11.11**
					Average of district	**5.00**	**33.05**	**40.83**	**26.29**

Note: F: Fixed plot; R: Random plot; 1st picking: 04/12/2014; 2nd picking: 19/12/2014 and 3rd picking: 03/01/2015

on the activity of mirid bugs/plant were recorded at weekly interval from the 20 randomly selected plants from each plot. For recording incidence of pink bollworm, 20 plants were randomly selected from the each plot and from each selected plant toal number of healthy squares and green bolls and total number of damaged squares and green bolls were counted at weekly interval and per cent incidence was worked out.

The activity of mirid bug was observed during 2nd week of November, 2014 to 4th week of January, 2015 in Kheda district. The population of this pest was ranged from 0.03 to 0.81/plant under monitoring fields. In Vadodara district, the activity of this emerging pest was not observed.

The activity of pink bollworm was observed during 2nd week of November, 2014 to 3rd week of February, 2015. The infestation on square due to pink bollworm was observed in the range of 0.04 to 17.70 per cent and 0.09 to 8.84 per cent in Vadodara and Kheda district, respectively. The highest (17.70 %) infestation on square due to pink bollworm was observed during 5th week of January, 2015 in Vadodara district whereas in case of Kheda district, it was during 3rd week of January, 2015. The infestation on green bolls due to pink bollworm was observed in the range of 2.50 to 47.79 per cent and 0.05 to 1.90 per cent in Vadodara and Kheda district, respectively. The highest (47.79%) infestation on green bolls due to pink bollworm was observed during 3rd week of February, 2015 in Vadodara district whereas in case of Kheda district, it was during 3rd week of January, 2015. As far as catches of *P. gossypiella* in pheromone traps (Table 2) is concerned, it was started from 2nd week of November, 2014 to 3rd week of February, 2015 to the tune of 0.11 to 4.33 moths/trap/week and 0.06 to 2.33 moths/trap/week in Vadodara and Kheda district, respectively. The peak activity of the pest was recorded during 4th week of December, 2014 *i.e.* more than 4 moths per trap in Vadodara district.

The monitoring of pink bollworm resistance was also worked out. For the purpose, boll samples (30 bolls/plot/village) from each selected villages were collected at 90, 105 and 120 days after sowing (DAS) and brought to the laboratory and worked out the per cent incidence by "Boll Destruction Method". The incidence of pink bollworm was high in Vadodara district (upto 94%) irrespective of the *Bt* cotton varieties under studies. In Vadodara district, the highest pink bollworm population was recorded in Karjan taluka (45.55%) followed by Dabhoi (19.44 per cent). The least incidence of pink bollworm was recorded in Savli taluka (11.11%) (Table 3). In case of Kheda district, incidence of pink bollworm was lower (upto 27%) as compared to Vadodara district irrespective of the *Bt* cotton varieties under studies. In Kheda district, the highest pink bollworm population was recorded in Kathalal taluka (10.55%) followed by Thasra (5.83%). The least incidence of pink bollworm was recorded in Kapadwanj taluka (2.22%) (Table 4). On an average, incidence of pink bollworm in *Bt* cotton in Vadodara and Kheda district was 26.29 and 5.52 per cent, repectively. Comparatively lower incidence of pink bollworm was recorded in Kheda district than Vadodara district. This variation could be due to area under cotton, initiation of pest activity and general sowing pattern for which vadodara has an edge over Kheda distrcit.

The field survival of pink bollworm revealed the possibility of resistance build up and farmers must have to resort appropriate resistance management strategies to sustain the BG technology especially in the hot spot areas. The emerging and future insect pests problem have to be tackled with IPM approaches

Table 4. Infestation of pink bollworm, *P. gossypiella* in *Bt* cotton in Kheda district during 2014 - 2015 (Boll Destruction Method)

Taluka	Village	Field number	Number of damaged bolls out of 30			Picking wise infestation of pink bollworm (%)			
			1st	2nd	3rd	1st	2nd	3rd	**Average**
Kapadvanj		F1	00	01	02	0	0	6.66	**2.22**
	Kapadivav	F2	00	00	01	0	0	3.33	**1.11**
		R1	00	00	03	0	0	10.00	**3.33**
		R2	00	01	00	0	0	0	**0.00**
					Average	**0.00**	**0.00**	**5.00**	**1.67**
	Antisar	F1	00	01	01	0	0	3.33	**1.11**
		F2	00	02	00	20.00	3.33	0	**7.78**
		R1	06	00	00	16.66	0	0	**5.55**
		R2	05	02	00	0	3.33	0	**1.11**
					Average	**9.17**	**1.67**	**0.83**	**3.89**
	Thavad	F1	00	02	01	3.33	6.66	3.33	**4.44**
		F2	00	00	02	0	0	6.66	**2.22**
		R1	00	01	00	0	0	6.66	**2.22**
		R2	00	01	01	0	0	0	**0.00**
Kathalal					**Average**	**0.83**	**1.67**	**4.16**	**2.22**
	Sikandar	F1	05	00	06	16.66	0	13.33	**10.00**
	porda	F2	00	01	03	0	3.33	10.00	**4.44**
		R1	03	04	01	10.00	13.33	3.33	**8.89**
		R2	09	02	00	30.00	6.66	0	**12.22**
					Average	**14.17**	**5.83**	**6.67**	**8.89**
	Vishwanath	F1	04	04	05	13.33	13.33	16.66	**14.44**
	pura	F2	00	02	01	0	6.66	3.33	**3.33**
		R1	03	00	00	10.00	0	0	**3.33**
		R2	00	00	04	0	0	13.33	**4.44**
					Average	**5.83**	**5.00**	**8.33**	**6.39**
	Continue								
	Laxmipura	F1	00	02	01	0	6.66	3.33	**3.33**
		F2	06	01	05	20.00	3.33	16.66	**13.33**
		R1	02	00	08	6.66	0	26.66	**11.11**
		R2	03	04	06	10.00	13.33	20.00	**14.44**
					Average	**9.17**	**5.83**	**16.66**	**10.55**
ThasaraLabhapura		F1	00	02	03	0	6.66	10.00	**5.55**
		F2	00	01	01	0	3.33	3.33	**2.22**
		R1	00	03	05	0	10.00	16.66	**8.89**
		R2	00	00	01	0	0	3.33	**1.11**
					Average	**0.00**	**5.00**	**8.33**	**4.44**
	Ajupura	F1	00	05	02	0	16.66	6.66	**7.77**
		F2	00	02	05	0	6.66	16.66	**7.77**
		R1	00	01	02	0	3.33	6.66	**3.33**
		R2	04	00	00	13.33	0	0	**4.44**
					Average	**3.33**	**6.66**	**7.50**	**5.83**
	Mugatpura	F1	03	02	02	10	6.66	6.66	**7.77**
		F2	00	00	07	0	0	23.33	**7.78**
		R1	00	00	03	0	0	10.00	**3.33**
		R2	02	02	00	6.66	6.66	0	**4.44**
					Average	**4.17**	**3.33**	**10.00**	**5.83**
					Average of district	**5.18**	**3.88**	**7.49**	**5.52**

Note: F: Fixed plot; R: Random plot; 1st picking: 04/12/2014; 2nd picking: 19/12/2014 and 3rd picking: 03/01/2015

as a part of a sustainable crop production technology.

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A rapid process for preparation of bio enriched compost from cotton stalks

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ABSTRACT : A rapid composting process was developed for the preparation of bio-enriched compost from cotton stalks. The cotton stalks were treated with microbial consortia to fasten the composting process. Large scale composting experiments with ten tonnes of chipped cotton stalks per trial were conducted at Nagpur, Maharashtra; Sirsa, Haryana and Coimbatore, Tamil Nadu during 2011 and 2013. The raw cotton stalks used for composting at Nagpur and Coimbatore regions were dried (< 15% moisture) while at Sirsa, the stalks were wet (> 30% moisture). There were two treatments viz., microbial consortia treated and untreated (control). In each treatment, three heaps of 1.7 tonnes each were made. After thirty days of initiation (DAI) of composting process (cooling phase), the treated compost was enriched with plant growth promoting microorganisms. The results showed the carbon: nitrogen (CN) ratio was reached near to 20 at 60 and 45 DAI of composting in dry and wet cotton stalks respectively while, in control (untreated), the compost was ready at 90 and 60 DAI of composting. Thus, 30 and 15 days could be saved by using microbial consortia for the preparation of bio-enriched compost from dry and wet cotton stalks respectively. The NPK content of bio-enriched compost was about three times higher than farmyard manure. Thus, the preparation of bio-enriched compost from cotton stalks would be a viable solution for *in situ* management of cotton stalks for increasing soil fertility.

Key words : CN ratio, composting, cotton stalks, microbial consortia, nitrogen, phosphorus, potassium

In India about 30 million tonnes of cotton stalks are generated every year (Hiloidhari *et al.,* 2014). Cotton stalks comprise about 75-82 per cent holocellulose, 24-26 per cent lignin, 12-14 per cent moisture and 6-8 per cent ether extractive. Harvested stalks stored in the field serve as storehouse for harmful insects and diseases. Sometimes cotton stalks are mulched in the soil that requires heavy machinery & some additional fertilizer need to be added to enhance the stalk decaying process, which further involves high energy and cost. Most of the cotton stalks are treated as waste, some are being used as fuel by rural population, while annually, and the bulk of the cotton stalk is burnt after harvest to clean the field, which then leads to nutrient losses and increased CO_2 inputs to the atmosphere. Hence the best way to bring back the nutrient into the soil is composting of cotton stalks.

Composting is the process in which complex molecules such as lignin, cellulose, hemicelluloses, lipids is converted into simpler molecules (Nagarajan *et al.,* 1985 and Summerel and Berges, 1989). During the composting process, besides the final product in the form of humus; heat, compounds of nitrogen, oxygen, phosphorus, CO_2, H_2O, and a significant amount of microbial biomass is created (Tiquia *et al.,*2002). Many factors like temperature,

moisture content, oxygen concentration and nutrient availability affects the rate of decomposition of organic matter. These factors, in turn, strongly affect the structure and diversity of the microbial community, microbial activities and the physical and chemical characteristics of the compost (Miller, 1993). Although considerable work has been done on composting of organic waste, composting of high lignin content organic material like cotton stalks within shorter period of time is still a challenge.

The moisture content of cotton stalks immediately after uprooting was 40 – 50 per cent and same was found drop from 50 to below 20 per cent when the stalks were left in the field, after uprooting for three weeks (Gemtos and Tsiricoglou, 1999). The cotton stalks are normally uprooted in central and southern parts of country when the plant is almost dry and the chipping of the uprooted dry cotton stalks left with a moisture of around 12 per cent (Anonymous, 2010). Hence, the response of these raw cotton stalks with varied moisture content towards composting in not yet known. Considering this, the present paper was aimed to study the influence of microbial consortia on preparation of bioenriched compost from wet and dry cotton stalks within shorter period and to characterize the prepared compost at different intervals.

MATERIALS AND METHODS

Microbial consortia : Microbial consortia consisted of aerobic consortium, anaerobic consortium and plant growth promoting microbes. The liquid aerobic and anaerobic microbial consortia were obtained from Microbiology lab, Central Institute for Research on Cotton Technology (CIRCOT), Mumbai. The microbial strains viz., *Bacillus stearothermophilus, Pleurotus flabellatus and Phanerochaete chrysosporium* were used for the preparation of aerobic consortium. The anaerobic consortium consisted of mixture of anaerobic and facultative anaerobic microbes. The commercial solid formulations of plant growth promoting microorganism's *viz., Azosprillum, Azotobacter, Fluorescent Pseudomonas, Phospobacteria and Trichoderma viridie* were obtained from IARI, New Delhi.

Cotton stalks : The cotton stalks were collected at Nagpur, Maharashtra; Sirsa, Haryana and Coimbatore, Tamil Nadu. The chipped cotton stalks of 3 to 4 cm length and 1 to 2 cm thickness were used for composting purpose. The chipped cotton stalks collected from Sirsa was termed as wet cotton stalks since the initial moisture recorded was 30-35 per cent while cotton stalks collected from Nagpur and Coimbatore were termed as dry cotton stalks since the moisture content recorded was 10 – 15 per cent.

Composting trial : The large scale trials on composting of cotton stalks using ten tonnes of chipped cotton stalks were conducted at CIRCOT unit at Nagpur, CIRCOT unit at Sirsa and Central Institute for Cotton Research (CICR) unit at Coimbatore respectively. There were six experiments conducted with two trials at each centre. The month and year of composting experiments carried out is indicated in Table 1. In each experiment, six heaps were made of 1.7 tonnes each. Three heaps were kept as control (without microbial consortia) and remaining three heaps were kept as treated (with microbial consortia). The ingredients *viz.,* alkali (0.2%), cattle dung (10%), garden soil (0.1), Urea (1.2%), Diammonium phosphate (2%), aerobic and anaerobic culture (0.1%) each in treated heaps only) were added and mixed in sequential

manner and heaps were made. The initial moisture content maintained was 50 per cent including the moisture content of raw cotton stalks. The individual heaps were covered with polythene sheets. At 30th day of composting, the plant growth promoting microorganisms (*Azosprillum, Azotobacter, Fluorescent Pseudomonas, Phospobacteria and Trichoderma viridie*) were added each @ 0.1% in the treated heap only. All the heaps were turned periodically for every week for proper aeration.

Table 1. Details of composting experiments

S. No.	Place	Month	Year
1.	CIRCOT unit, Nagpur	June - August	Trial I (2011), Trial II (2012)
2.	CIRCOT unit, Sirsa	January - March	Trial I (2012), Trial II (2013)
3.	CICR unit, Coimbatore	June - August	Trial I (2011), Trial II (2013)

Analysis of compost samples : The samples were taken from each heap at periodic intervals at 0, 15, 30, 45, 60 and 90 days of composting, oven dried and powdered. The powdered samples were passed through 200 µ size sieve and used for analysis. Each sample was analyzed for organic carbon (Walkey and Black, 1934), Total N by micro-Kjeldhal (Humphires, 1956), Total P (Jackson, 1973), Total K by Flame photometer (Stanford and English, 1949) and Cellulose, Hemicellulose and Lignin (Van Soest, 1963). CN ratio was determined by finding out ratio between total organic carbon and total nitrogen.

The fresh samples taken during different intervals were used for analyzing pH, enzyme activity and total microbial count. The pH was measured using pH meter. The cellulase activity was determined according to Benefield, 1971. The urease activity was measured by following the method of Tabatabai and Bremner, 1972. The unit of cellulase activity was expressed in mg of glucose released/g of sample / h while urease activity was expressed in mg of ammonium N released /g of sample / h. The total microbial count including total bacterial, total fungi and total actinomycetes counts were determined using 1g of wet sample by standard serial dilution technique.

RESULTS AND DISCUSSION

Use of organic wastes in agriculture is known to reduce the pollution and improve soil quality. An attempt was made in the present study to utilize the cotton stalks, an organic waste, which is locally available in large quantities after harvesting and having the disposal problem in major cotton growing states of India. Farmers are burning the stalks obtained after harvesting as they take more time for biodegradation due to its high lignin content and broad C: N ratio. At present, the cotton stalks are mostly burnt and sometimes used as domestic fuel. Hence, an attempt was made to accelerate the process of composting of cotton stalks using efficient lignin degrading microbes and enrich the compost with plant growth promoting microorganisms.

The microbial consortia include aerobic consortium, anaerobic consortium and plant growth promoting microorganisms. The trial was taken for ten tonnes of chipped cotton stalks and the compost samples were analyzed for various properties. The results on compost prepared from wet cotton stalks were reported based on the consolidation of trial results taken at Sirsa. The results on compost prepared from dry cotton stalks were reported based on the consolidation of trial results taken at Nagpur and Coimbatore.

During the composting process, there was an increase in temperature in compost heaps was recorded from 10 - 15 DAI and the trend was prolonged up to 30 to 35 DAI. The maximum temperature recorded in the compost heaps during the thermophilic phase was 55 - 60°C (data not shown). The mineralization occurs most rapidly during the thermophilic phase of composting (40 – 60° C) which lasts for several weeks or months depending on size of the system and composition of the ingredients. High temperature (60-75°C) reached in thermophilic phase of composting is a factor which completely reduces the number of pathogens (Macgregor *et al.,* 1981).

CN ratio : CN ratio is an important indicator for composting process. As per Fertilizer Control Order (FCO), Government of India, 1985 (FAI, 2007), the CN ratio should reach below 20 to indicate the process of completion of composting. In the present study, the changes in CN ratio for wet and dry cotton stalks during different intervals were evaluated. The composting experiments conducted using wet cotton stalks at Sirsa during the period 2011 and 2012 showed that the CN ratio of treated wet cotton stalks reached to 20.0 at 45 DAI while untreated wet cotton stalks reached to 20.0 at 60 DAI (Fig. 1). Thus, the results indicated that the treatment of cotton stalks with microbial consortia could reduce the time period of preparation of compost from wet cotton stalks by fifteen days. Similarly, the composting experiments conducted during 2011 and 2013 using dry cotton stalks at Coimbatore and Nagpur suggested that CN ratio reached to 20.0 at 60 DAI in treated and 90 DAI in untreated compost respectively. The results showed the treatment of microbial consortia reduce the time period of preparation of compost from dry cotton stalks by thirty days. CN ratio is an important indicator for composting process. Several workers have suggested that inoculation of fungal cultures resulted in rapid decomposition of agro-residues with decreases in CN ratio (Jagadeesh *et al.,* 1996 and Singh *et al.*, 1992).

In this study, to accelerate the composting process, mixture of lignocelluloytic microorganisms including *P. chrysosporium*, *B. stearothermophilus* and *P. flabellatus* was used. In a similar study, the inoculation of lignocellulolytic microorganisms such as *P. sajor-caju*, *T. harzianum*, *A. niger* and *Azotobacter chrococcum* to enhance the composting process was reported for wheat straw composting (Singh and Sharma, 2002). The composted cotton stalks had significantly reduced hemicelluloses and lignin than cellulose when compared to raw cotton stalks (results not shown). The observed results might be due to more susceptibility of hemicelluloses towards microbial degradation than other counterparts. During the degradation, part of lignin is also degraded which results in decrease in lignin content. Similar results were found by Kadalli, 1999 and Nagarajan *et al.*, 1985. The fungal pretreatment of cotton stalks by *P. chrysosporium* showed significant lignin and hemicelluloses degradation compared to untreated stalks (Shi *et al.,* 2009). The lignocellulosic degradation during composting process was due to composite cultures than single organism in which each organism have different specificity and thus brings faster decomposition (Arora and Garg, 1992 and Gupta *et al.,* 2004).

Biological characterization : The samples taken during different intervals of composting were analysed for biological characteristics such as enzyme activity and total microbial count. The enzyme activities such as

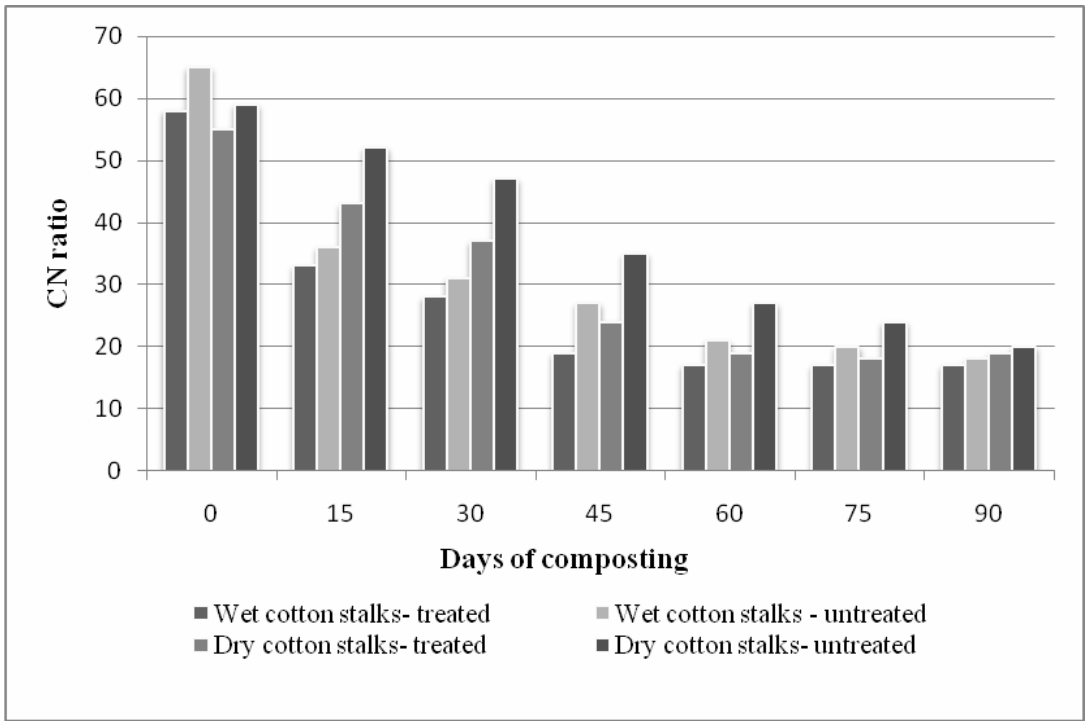

Fig. 1. CN ratio of compost prepared from cotton stalks during different intervals

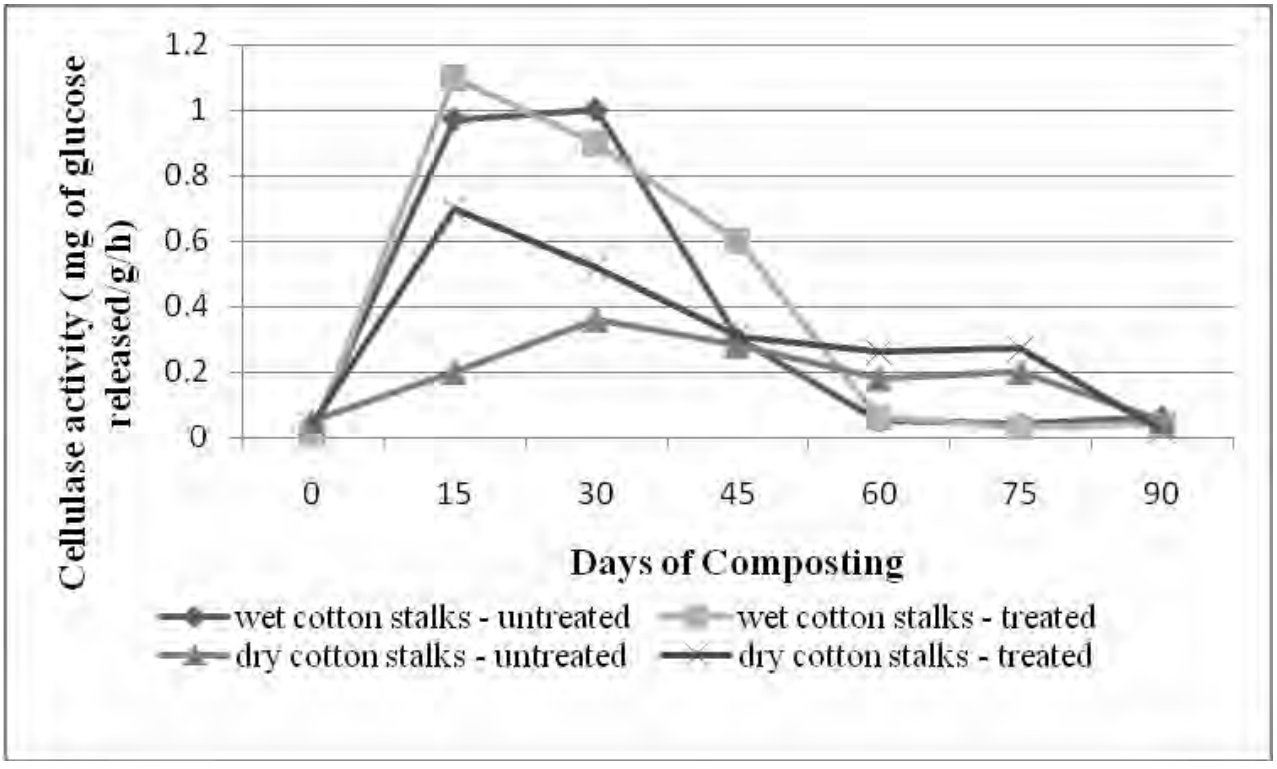

Fig. 2. Cellulase activity of compost prepared from cotton stalks during different intervals

Table 2 Total microbial count of compost prepared from cotton stalks during different intervals

Treatments	DAI of composting											
	Total bacterial count (cfu x 10^9)				Total fungi count (cfu x 10^5)				Total actinomycetes count (cfu x 10^7)			
	0	30	60	90	0	30	60	90	0	30	60	90
Wet cotton stalks- untreated	1.0^{c}	2.6^{b}	3.3^{b}	4.1^{a}	3.5^{b}	4.4^{ab}	2.4^{b}	1.7^{c}	4.0^{b}	20.0^{b}	36.0^{*}	30.0^{*}
Wet cotton stalks - treated	2.8^{b}	45.0^{a}	4.0^{b}	5.2^{a}	110.0^{a}	4.8^{a}	3.9^{b}	2.4^{bc}	9.7^{b}	28.0^{ab}	48.0^{*}	38.0^{*}
Dry cotton stalks - untreated	2.7^{b}	3.8^{b}	31.0^{a}	4.5^{a}	2.8^{b}	1.5^{c}	11.0^{ab}	3.6^{b}	9.6^{b}	17.0^{b}	36.0^{*}	47.0^{*}
Dry cotton stalks - treated	8.0^{a}	32.0^{a}	48.0^{a}	12.0^{b}	130.0^{a}	3.2^{b}	19.0^{a}	5.7^{a}	20.0^{a}	37.0^{a}	45.0^{*}	50.0^{*}

DAI: Days after initiation; * Non Significant; Treatment values followed by same alphabet do not differ significantly. There were three replications per treatment. The values are the means of two different experiments.

cellulase and urease activity were studied. The cellulase activity (mg of glucose released/g/h) was found to be higher in 15 days after initiation (DAI) of composting in the microbial consortia treated cotton stalks (Fig. 2). The cellulase activity recorded was 1.1 and 0.7 in treated wet and dry cotton stalks respectively while in the untreated cotton stalks, the cellulase activity was higher during 30th DAI. The cellulase activity recorded in untreated wet and dry cotton stalks was 1.0 and 0.4, respectively. The cellulase activity was recorded higher during the initial stages of composting process and drastically reduced after 60th DAI. In a similar study, Semenov *et al.,* 1995 reported that cellulase activity was increased with increase in temperature. The urease activity (mg of ammonium N released/ g/h) was found to be 0.08 at 0th DAI in dry and wet cotton stalks in treated as well as untreated. During the remaining period of composting, the urease activity was found in the range of 0.02 to 0.04 and there was no difference in the activity among treated and untreated cotton stalks was observed (results not shown).

The total microbial count including bacteria, fungi and actinomycetes were estimated in compost at different intervals (0,

Table 3. Physico chemical characterization of compost prepared from wet cotton stalks during different intervals

DAI of composting		Physico chemical characters				
		pH	Organic carbon (%)	Total nitrogen (%)	Total phosphorus (%)	Total potassium (%)
0	UT	8.0^{a}	38.0^{a}	0.6^{b}	0.2^{e}	0.2^{g}
	T	7.8^{a}	33.0^{b}	0.6^{b}	0.2^{e}	0.3^{fg}
45	UT	7.3^{b}	28.0^{c}	1.0^{a}	0.6^{d}	0.5^{ef}
	T	7.1^{b}	22.2^{d}	1.1^{a}	0.9^{bc}	0.8^{bcd}
60	UT	7.1^{b}	21.0^{d}	1.0^{a}	0.8^{cd}	0.6^{de}
	T	7.1^{b}	20.4^{d}	1.1^{a}	1.1^{ab}	0.9^{abc}
75	UT	7.2^{b}	20.0^{d}	1.0^{a}	0.9^{b}	0.7^{cde}
	T	7.2^{b}	18.5^{d}	1.1^{a}	1.2^{a}	0.9^{abc}
90	UT	7.1^{b}	20.0^{d}	1.1^{a}	0.9^{bc}	1.0^{ab}
	T	7.2^{b}	19.4^{d}	1.2^{a}	1.2^{a}	1.1^{a}

DAI: Days after initiation; UT: Untreated; T: Treated Treatment values followed by same alphabet do not differ significantly. There were three replications per treatment. The values are the means of two different experiments.

Table 4 Physico-chemical characterization of compost prepared from dry cotton stalks during different intervals

DAI of composting		Physico chemical characters				
		pH	Organic carbon (%)	Total nitrogen (%)	Total phosphorus (%)	Total potassium (%)
0	UT	8.0[a]	35.0[*]	0.6[d]	0.2[e]	0.8[c]
	T	7.8[ab]	33.0[*]	0.6[d]	0.2[e]	0.9[c]
45	UT	7.5[bc]	35.0[*]	1.0[cd]	0.3[de]	1.1[bc]
	T	7.1[c]	30.0[*]	1.2[bc]	0.5[cd]	1.1[bc]
60	UT	7.0[de]	35.0[*]	1.3[bc]	0.3[de]	1.3[abc]
	T	7.3[cd]	30.0[*]	1.6[ab]	0.8[ab]	1.5[ab]
75	UT	6.7[e]	34.0[*]	1.4[abc]	0.5[cd]	1.3[abc]
	T	6.9[e]	32.0[*]	1.8[a]	1.0[a]	1.8[a]
90	UT	6.8[e]	33.0[*]	1.6[ab]	0.6[bc]	1.5[ab]
	T	6.8[e]	34.0[*]	1.8[a]	0.9[a]	1.8[a]

DAI: Days after initiation; UT: Untreated; T: Treated; [*] Non Significant; Treatment values followed by same alphabet do not differ significantly. There were three replications per treatment. The values are the means of two different experiments.

15, 30, 45, 60 and 90 DAI). The enumerated microbial count at 0, 30, 60 and 90 DAI is presented in Table 2. The total bacterial count was found higher in compost prepared from treated wet and dry cotton stalks at 30th DAI and 60th DAI respectively. The respective values were 45× 10^9 cfu/g and 48× 10^9 cfu/g. The total fungal count was higher at 0th DAI in both the wet and dry cotton stalks treated samples and their respective counts were 110 × 10^5 cfu/g and 130 × 10^5 cfu/g. The total actinomycetes population was higher during the later stage of composting (60th and 90th DAI). The actinomycetes population during this period did not differ significantly among the treatments. Among the microbial populations, bacterial population dominated all stages of decomposition process followed by actinomycetes and fungi. The similar results were obtained by Nodar *et al.,* 1990. It was obvious that increase in microbial population in treated compost than untreated was due to addition of microbial consortia including plant growth promoting microorganisms in the treated compost.

Physico chemical characterization : The physico-chemical properties of compost prepared from wet and dry cotton stalks are presented in Table 3 and Table 4, respectively. The initial pH of the compost at 0th DAI was slightly alkaline 8.0 ± 0.2. After 45th DAI, the pH of the compost was in the range of 7.0 ± 0.5. At 0th DAI of composting, the organic carbon (OC) content (%) was in the range of 33 to 38% (Table 3 and 4). The OC (%) content was reduced from 30 to 20 and 33 to 19.4 in untreated and treated wet cotton stalks compost respectively (Table 3) while the OC was unchanged in dry cotton stalks composting which needs further investigation (Table 4). The results are in accordance with the observations of Gupta *et al.,* 2004; Imam and Sharanappa, 2002 who also reported decrease in the organic carbon when different crop residues was composted with poultry manure. In a similar study, the treatment of cotton stalks with *P. chrysosporium* and *Azotobacter* resulted in decrease in OC (%) from 43.9 to 30.65 and increase in TN (%) from 1.46 to 2.16 was observed over the period of 16 weeks (Seoudi, 2013). The

total nitrogen (%) (TN), total phosphorus (%) (TP) and total potassium (%) (TK) was recorded higher in treated cotton stalks than untreated cotton stalks. As stated earlier (Fig. 1), the CN ratio was reached to 20 at 45 DAI and 60 DAI in microbial consortia treated wet and dry cotton stalks respectively. Similarly, significant increase in TN, TP and TK contents were observed at 45 DAI and 60 DAI in microbial consortia treated wet and dry cotton stalks respectively. The TN, TP, TK contents recorded in treated wet cotton stalks compost at 45 DAI and 90 DAI of composting were 1.1, 0.9 0.8 and 1.2, 1.2, 1.1 respectively (Table 3). The TN, TP, TK contents was higher in compost prepared from dry cotton stalks than wet cotton stalks. The TN, TP, TK contents recorded in treated dry cotton stalks compost at 60 DAI and 90 DAI of composting were 1.6, 0.8, 1.5 and 1.8, 0.9, 1.8, respectively.

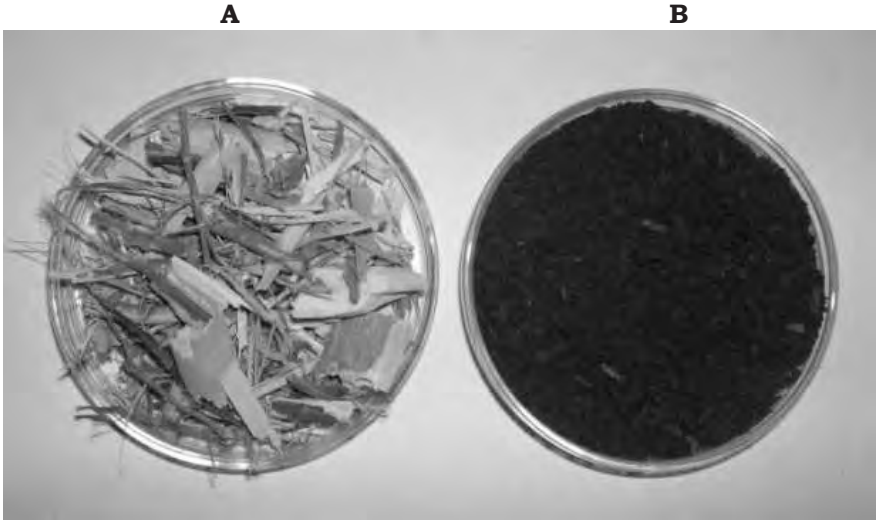

Fig. 3. Bio-enriched compost from cotton stalks
(A- raw cotton stalks; B- bio-enriched cotton stalks compost)

The increase in nitrogen content during composting might be a direct manifestation of mass carbon loss (Nagarajan *et al.*, 1985; Maleena, 1998; Singh *et al.*, 1992). Similar results were obtained by Gaur and Singh, 1982 that there was 27 per cent increase in nitrogen content, when mechanized compost inoculated with *Azotobacter* and rock phosphate. It is also evident from the experiments of Kapoor et al., 1983 that *Azotobacter* inoculation helps in increasing the N content of compost. It has also been reported by Kavitha and Subramanian, 2007 that the phosphorus content was increased conspicuously with addition of composted poultry litter, rock phosphate and microbial inoculants. The FCO recommends that the TN, TP and TK content (%) in the compost should be 1.0, 0.8 and 0.8 respectively and colour of the compost should be dark brown to black (FAI, 2007). Our results were in accordance with FCO where the colour of the compost was completely turned to black. The matured bio-enriched compost prepared from wet cotton stalks is presented in the Fig. 3. Thus these results indicated that physico-chemical properties of the microbial consortia treated compost prepared from wet and

dry cotton stalks at 45 DAI and 60 DAI respectively meets the quality standards as prescribed by FCO. The TN, TP, TK content (%) of farm yard manure (FYM) was calculated to be 0.5, 0.2, 0.5 respectively (results not shown). It was noted that the TN, TP, TK level of bio-enriched compost prepared from dry cotton stalks in this study was about three times higher than FYM.

CONCLUSION

In the present paper, the effect of microbial consortia on rapid composting and nutrient level improvement in wet and dry cotton stalks were studied. The results showed that, good quality compost was prepared at 45 and 60 days from wet and dry cotton stalks using microbial consortia, while the same was obtained at 60 and 90 days respectively without microbial consortia. Thus fifteen and thirty days could be saved for the preparation of compost from wet and dry cotton stalks respectively. The results also showed that the nutrient level of microbial treated compost is better than untreated compost. Thus the process developed will not only yield good quality compost but also a viable on-farm solution for effective management of cotton stalks. In future, experiments may be conducted for mechanization of this process for large scale preparation of compost from cotton stalks at farm level.

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Enhancement of UV protective property of cotton fabric by using plant extract

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ABSTRACT : The present study have been conducted to enhance the UV protective property of cotton fabric and to assess its acceptability by consumers. *Syzygium cumini* (jamun) plant was selected for this study and phytochemical analysis was conducted. Methanolic leaves extract of *Syzygium cumini* was prepared with soxhlet extraction method. The cotton fabric was treated with *Syzygium cumini* leaves extract by using pad-dry-cure process and was evaluated for ultraviolet protection factor (UPF). Results of phytochemical analysis revealed the presence of tannin, flavonoids, phenols which possess good UV absorbing properties as reported in review. The cotton fabric when treated with *Syzygium cumini* leaves extract exhibited 29.5 UPF value, providing very good protection as compared to untreated cotton fabric which exhibited 10.9 UPF value, providing poor protection. The results of the acceptability of developed cotton fabric revealed; majority of the respondents, preferred the developed cotton fabric with and without surface enrichment, for female garments followed by male and children garments. It was concluded that by improving UV protective property of cotton fabric using natural source such as plants can provide safe and healthy life for each member of the society and can increase the value and demand of cotton based textiles.

Keywords: Cotton fabric, syzygium cumini, phytochemical analysis, UV protection

Cotton fiber is amazingly versatile, whether alone or blended, it out shells all other fibers combined. Consumers know that fabrics made from cotton put forth natural comfort, visual appeal, durability and value. With the mercury scorching up in summer time, people prefer to hide behind cotton fabric due to its inherent properties like good absorbency, ecofriendly nature, lightweight and many more. According to the *Lifestyle and Retail Monitor*™ Survey, more than 9 in 10 (almost 100 percent) consumers state that they would like to choose cotton over synthetic active wear if cotton could wick moisture, regulate temperature, be lightweight, hold or lock color, resist UV rays (Cotton Incorporated Lifestyle Monitor, 2015). Due to depletion of ozone layer and high exposure to UV rays, problems such as sunburn, premature skin ageing, skin cancer and eye disorders are increasing (Holme, 2003). Clothing provides one of the most convenient forms of protection against UVR but not all type of garments provides sufficient sun protection. (http:www.arpansa.gov.au/services.unf/index.gfm). Studies reveal that favourite fibers like cotton, rayon, flax are the poorest UV absorbers, as compared to polyester, wool, silk and nylon (Crews *et al.*, 1999). Chemicals, currently used to impart ultraviolet radiation protection property to textile materials, are titanium di-oxide, zinc oxide along with the organic ultraviolet radiation absorbers such as phenylbenzotriazole and dibenzoylmethanes (Subtamaniyan *et al.,* 2013). Many such chemicals commonly used have not been established safe for long term human use.

Plant extracts and their active metabolites possess several types of activities such as antibacterial, antifungal, antioxidant, antiviral, etc. *Syzygium cumini* is one of the most important medicinal plant belonging to Myrtaceae family and is commonly known as jamun. In the traditional medicine systems, different parts such as seeds, leaves of *Syzygium cumini* have been used as liver tonic, to strengthen teeth and gums, diabeties, treat leucorrhoea, stomachalgia, fever (Soni *et al.,* 2011). Maske *et al.,* 2012 in their study, reported *Syzygium cumini* as an effective sunscreen agent for protection of skin from harmful ultraviolet radiations. Recognizing the importance of cotton in our life and consumer requirements for healthy life, this study have been conducted to develop UV protective fabric by using a natural source (*Syzgium cumini)* which has not been explored till date for its UV protective property on cotton fabrics.

MATERIALS AND METHODS

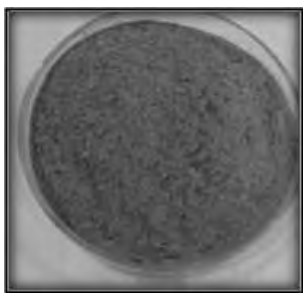
Material used : Fresh and mature leaves of *Syzygium cumini* (jamun) were collected from CCS, Haryana Agricultural University, Hisar. Various chemicals/auxiliaries such as methanol (solvent), Americos Amylase 543 (desizing agent), Palkascour (scouring agent), Fixa prêt Eco (resin cross linking agent), Magnesium chloride (catalyst) and 100 per cent plain woven cotton fabric were procured from the respective industries and the preliminary data of the procured fabric is presented in Table 1.

Table 1. Preliminary data of cotton fabric

Parameters Fabrics	Fabric count (ends and picks/ sq. inch)		Fabric thickness (mm)	Fabric weight (gm/m^2)
	Warp	Weft		
Woven	95	76	0.233	119

Preperation of *Syzygium cumini* leaves extract : 40gms of fresh and mature leaves of *Syzygium cumini* (jamun) were collected, washed and shade dried. The dried leaves were grounded in powder form and subjected to hot methanolic extraction at 55-60^0 C by using soxhlet extraction process, as described by Choudhary *et al.,* 2012 with slight modification.

Table 2. Steps followed for preperation of Syzygium cumini leaves extract

Plant	Fresh leaves	Dry leaves	Powdered leaves	Soxhlet extraction
Syzygium cumini				

Preperation of cotton fabric : The greige cotton fabric was treated with 3 per cent concentration of alpha amylase with 1:50 material to liquor ratio at 60^0C for 45 minutes reaction time, at pH 6-7 (Vigneswaran *et al.,* 2013). The desized cotton fabric was further subjected to enzymatic scouring with 2 per cent enzyme concentration with 1:50 material to liquor ratio for 45 minutes duration at 40^0 C, pH 8. After the treatment, the fabric was rinsed thoroughly, first with hot water and then with cold water to neutralize the enzymatic effect (Ragendran *et al.,* 2011).

Qualitative phytochemical analysis : Freshly prepared extract of the powdered leaves was subjected to phytochemical analysis to find the presence of flavonoids, tannins, phenols according to the procedure described by Gopinath SM *et al.,* 2012; Saidulu Ch *et al.,* 2014).

1. **Detection of tannins**
   Ferric chloride test: To the filtrates, a few drops of ferric chloride solution were added. A blackish precipitate indicates the presence of tannins.
2. **Detection of flavonoids**
   Lead acetate test: Extracts were treated with few drops of lead acetate solution. Formation of yellow color precipitate indicates the presence of flavonoids.
3. **Detection of phenolics compounds**
   Ferric chloride test : Extracts were treated with 3-4 drops of ferric chloride solution. Formation of bluish black color indicates the presence of phenols.

Application of syzygium cumini leaves extract:

Pad dry cure process : To develop Ultraviolet protective fabric, cotton fabrics were treated with *Syzygium cumini* leaves extract in the presence of resin cross linking agent and magnesium chloride by using pad-dry-cure process. The sample was immersed in finishing solution containing 3 per cent (owf) concentration of methanolic leaves extract of *Syzygium cumini,* with 1:20 material to liquor ratio, 50g/l resin cross linking agent (Fixa prêt eco), 10g/l magnesium chloride, at 5pH for 30 minutes. The fabric was then passed through the padded roller on two-bowl pneumatic padding mangle at a pressure of $2kg/cm^2$ with two dips and nips. The fabric was dried and cured in laboratory curing chamber.

Quantitative assessment of ultraviolet protection factor (UPF) : The ultraviolet protective property of cotton fabric treated with *Syzygium cumini* leaves extract in presence of resin cross linking agent and magnesium chloride was evaluated by using UVR TRANSMISSION AATCC-183:2004 test method. The transmission of ultraviolet radiation (UV-R) was determined in the wavelength range of 280-400nm by using Compsec M 350 UV-Visible Spectrophotometer. UVA and UVB percentage transmission were measured. Ultraviolet protection factor was calculated using mean percentage transmission in UVA region (320-400 nm) UVB region (280 -320 nm) according to the following equation:

$$UPF = \frac{\sum_{\lambda 290}^{400} E\lambda \, xS\lambda \, x\Delta\lambda}{\sum_{\lambda 290}^{400} E\lambda \, x \, S\lambda \, x \, T\lambda \, x\Delta\lambda}$$

Where:
Eë = relative erythermal spectral effectiveness
Së= solar spectral irradiance
Të= average spectral transmission of the specimen
[30]%ë= measured wavelength interval (nm)

Table 3. Grades and Classification of UPF

UPF Range	UVR transmission (%)	Protection category
15 to 24	5.0-2.4	Good protection
25 to 39	3.3-2.5	Very good protection
40 to 50	<2.5	Excellent protection

Assessment of developed UV protective cotton fabric by consumers : The assessment was conducted in Hisar city of Haryana state. Thirty respondents were purposively selected by taking care that the respondents should be female, married and should fall in the age group of 30-45, as females falling in this age group are more receptive towards buying and caring for the clothing needs of the family members such as children, husband, brother in law, mother in law. Questionnaire was prepared for the collection of data regarding the awareness of respondents about the availability of UV protective garments in the market and their opinion regarding the developed UV protective cotton fabric for garment production.

RESULTS AND DISCUSSION

Plants are naturally gifted with number of medicinal compounds also known as phytochemicals. The phytochemicals are non-nutritive chemicals produced by plants for their own protection, but these have been found to protect humans against diseases as studied through recent researches. These includes tannin, alkaloid, flavonoids, and saponin (Edeoga *et al.,* 2005) having biological properties such as antioxidant activity, antimicrobial, stimulation of immune system, modulation of hormone metabolism, anticancer, UV protective and many more (Yadav and Agarwala, 2011). An experiment was conducted to evaluate the methanolic leaves extract of *Syzygium cumini* for the presence of tannin, flavonoids and phenols which have been reported to exhibit UV protective properties (Chaudhary and Mukhopadhay 2012; Sharma *et al.,* 2012). Phytochemical analysis revealed the presence of various secondary metabolites in the methanolic leaves extract of *Syzygium cumini* i.e tannin, flavonoids and phenols as shown in table 4. Thus due to the presence of such phytochemicals methanolic leaves extract of *Syzygium cumini* was applied on cotton fabric to develop and evaluate UV protective property of treated cotton fabric.

Ultraviolet Protection properties of treated cotton fabric : UPF is the scientific term used to indicate the amount of Ultraviolet (UV) protection provided to the skin by fabric. The higher the value, the longer a person can stay in the sun until the area of skin under the fabric becomes red (Dhandapani and Sarkar, 2007). According to experts, including the U.S. Environmental Protection Agency, the technique that best protects the skin against UV radiation is clothing (U.S. Environmental Protection, 2006). Fabrics with UPF values greater than 40 are considered as having excellent UV protection, whereas fabrics with 25-39 translates to have very good UV protection and fabrics with UPF values between 15-24 gives good UV protection as been indicated in table 3.

This study was conducted to develop the UV protective cotton fabric by using *Syzygium cumini* leaves extract and assess the acceptability of developed fabric. Table 5 shows the UPF values, UVA and UVB per cent transmission and UPF rating of the fabric under study. As indicated from the results, the treatment of cotton fabric with *Syzygium cumini* leaves extract improves the UV protection

Table 4. Phytochemical analysis of Syzygium cumini leaves extract

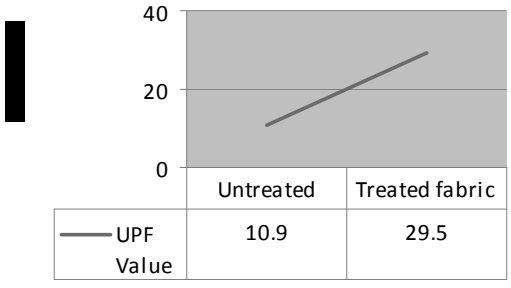
Phytochemical	Plants	Interface	Results
Tannin	*Syzygium cumini*	Blackish precipitates	Positive
Flavonoids	*Syzygium cumini*	Yellow color	Positive
Phenols	*Syzygium cumini*	Bluish black	Positive

property of the cotton fabric. The untreated cotton fabric indicates the UPF value of 10.9 exhibiting poor UV protection and treated cotton fabric exhibit UPF value of 29.5, providing very good UV protection. The UVA and UVB per cent transmission is also discussed as UVA and UVB radiation provides harmful effect on skin; UV-A can penetrate the top layer of skin, thereby damaging the inner layers. UV-B radiation can cause sunburn and is thought to be the major reason for skin cancer as it inhibits the synthesis of DNA, RNA and proteins (Holme, 2003). The data presented in table 5 and graph 2 shows the reduction in UVA and UVB percent

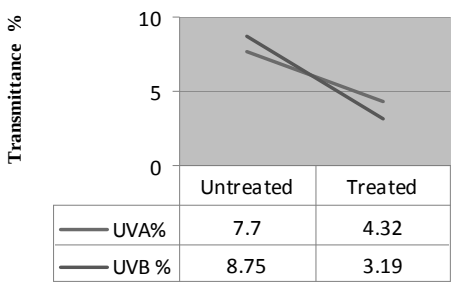
Table 5. Ultraviolet Protection Factor of the untreated and treated cotton fabrics

Fabrics	Ultraviolet Protection Factor (UPF)(AATCC-183:2004)				
	UVA(%)	UVB(%)	UPF Mean (value)	UPF range	Protection category
Untreated	7.70	8.57	10.9	No range	Poor
Treated	4.32	3.19	29.5	25-39	Very good

Graph:1 Ultraviolet protection value of cotton fabric

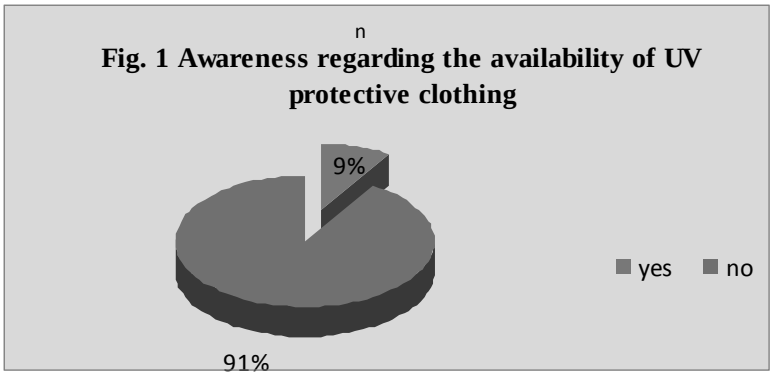


Graph:2 UVA and UVB percent transmitance of cotton fabric



transmission in cotton fabric, when treated with *Syzygium cumini* leaves extract, thus exhibiting higher UV protection.

The Ultraviolet protection provided by methanolic leaves extract of *Syzygium cumini* extract could be due to the presence of tannin, flavonoids and phenols. These are reported to protect plants from harmful UV rays as well as they are also good UV absorbers as reported by Subramaniyan *et al.,* 2013. Khazaeli and Mehrabani, 2008, analysed sun protective ability of some medicinal plants and observed the high



SPF value of V. tricolor and rendered such activity due to the presence of phenolics compounds and flavonoids. UV absorbers incorporated into fibers convert electronic excitation energy into thermal energy, function as radical scavengers and singlet oxygen quenchers. The high-energy, short wavelength ultraviolet radiations excite the UV absorbers to a higher energy state; the energy absorbed may then be dissipated as longer wavelength radiations (Das, 2010).

Assessment of developed UV protective cotton fabrics by consumers : This section deals with the awareness of the respondents regarding the availability of UV protective clothes/ garments in market as well as the acceptability of developed UV protective cotton fabric. Different type of garments such as Swin shirts, UV suits, sun suits and sun hats are manufactured by companies and are available in the market, which provide good UV protection (Das, 2010). In Fig. 1, it can be seen that only 9 per cent of the respondents were aware of the availability of UV protective clothes in market while 91 per cent of respondents were not aware. As results revealed, very less number of people are aware of the availability of such UV protective garments, there is a need to disseminate the information regarding the same among masses.

The data from Fig. 2 reveals the opinion of the respondents regarding the acceptability of developed (UV protective) cotton fabric. It was found that the majority of the respondents i.e 87 per cent preferred developed UV protective cotton fabric for female garments, 79 per cent for male garments and 84 per cent for children garments. The acceptability of developed cotton fabric may be attributed to the enhanced performance property of cotton fabric which increases its use in summers especially for outdoor activities. Respondent suggested that commonly used garments such as kurti, shirt, stoles, scarf, jumpsuit hats, caps can be developed by using the developed UV protective cotton textiles and used in day to day life.

The respondents were also shown surface

enriched developed UV protective cotton fabric by using block printing technique and asked for their opinion regarding the its acceptability for garments. The results as shown in Fig. 2 revealed that majority of the respondent i.e 95 per cent gave their acceptability for block printed developed UV protective cotton fabric for female garments followed by 37 per cent for male garments and 17 per cent for children garments. Hence it was cocludecd that as per the consu,er opinion block printed and plain were excepted for making garments.

Fig. 2 Opinion regading the acceptability of developed UV protective cotton fabric

	Female garments	Male garments	Children Garments
UVP cotton fabric	87	79	84
UVP cotton fabric with embelishment	95	37	17

UVP- Ultraviolet protective cotton fabric Multiple responses

CONCLUSION

Now –a-days with more environmental consciousness and more health problems associated with the incidence of skin cancer due to excessive exposure to sunlight as well as the use of synthetic products, consumers are looking for products with additional performance properties, developed through natural sources such as plants. Cotton being the most versatile and one of the favourite fiber for summer, can be treated with *Syzygium cumini* leaves extract which exhibits the potential to enhance its UV protective property thus providing a safe shield from harmful effect of UV radiations not only to outdoor workers but each member of the society when used as apparel.

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