STUDIES ON GENETIC INTROGRESSION IN COTTON THROUGH INTERSPECIFIC HYBRIDIZATION

Thesis submitted to the
University of Agricultural Sciences, Dharwad
in partial fulfillment of the requirements for the
Degree of

MASTER OF SCIENCE (AGRICULTURE)

In

GENETICS AND PLANT BREEDING

By
Ch. MADAN MOHAN

DEPARTMENT OF GENETICS AND PLANT BREEDING
COLLEGE OF AGRICULTURE, DHARWAD
UNIVERSITY OF AGRICULTURAL SCIENCES,
DHARWAD-580005

SEPTEMBER, 2005

ADVISORY COMMITTEE

Dharwad
AUGUST, 2005

(P.M. SALIMATH)
MAJOR ADVISOR

Approved by:

Chairman :______________________

(P.M. SAILMATH)

Members :1.____________________

(I.S. KATAGERI)

2.____________________

(H.M. VANADEVAIAH)

3.____________________

(B. FAKRUDDIN)

CONTENTS

LIST OF TABLES

LIST OF FIGURES

LIST OF PLATES

LIST OF APPENDICES

I. INTRODUCTION

Cotton, the 'White Gold' and 'King of fibre crops' enjoys a pre-eminent status among all the cash crops in the country and elsewhere by providing principle raw material, cotton for textile industry. It is an important agricultural commodity providing remunerative income to millions of farmers both in developed and developing countries. About 60 million people in our country are engaged in textile industry. In India, inspite of severe competition from synthetic fibres in recent years, it is occupying the premiere position with 70 per cent share in the textile industry.

Currently *Gossypium* includes 50 species, four of which are cultivated, 44 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 *Gossypium arboreum* L. are diploids (2n = 2x = 26) and are commonly called as old world cottons. It was one of the first crops to which, the rediscovered Mendelian principles were applied (Balls, 1906).

India has the distinction of having the largest cotton area (8.97 million ha) in the world with a production of 23.2 million bales and a productivity of 440 kg per ha (Anon., 2005a). India stands first in area, third in production and seventeenth in productivity with respect to cotton. Karnataka produces around 8 lakh bales of cotton lint from an area of 5.5 lakh ha with a productivity of 246 kg per ha (Anon., 2005b). Per capita availability of cloth has gone up from 22.65 sq. m. during 1989-90 to about 30 sq. m during 1999-2000 (Puri and Mehetre, 2001). The extrapolated requirement of 2050 A.D. with a projected population of 160 million is placed around 282 lakh bales of cotton production in India. Desi cotton occupies only 27 per cent of the total cotton area and the remaining area (73%) is under tetraploids.

Nearly one-third of India's export earnings are from textile sector where cotton alone constitutes nearly 70 per cent of raw material. No crop competes with cotton in respect of its potentiality for value added products.

India can take pride for being the only country growing a wide spectrum of quality cottons right from the shortest and coarsest, capable of spinning six counts to longest and finest used for producing 120 count yarns. Although, Indian cottons have very wide quality spectrum, the right combination of fibre length, micronaire and fibre strength is however absent in many of the popular varieties. The deficiency is particularly discernable in staple range 27 to 30 mm combined with micronaire of 4.0 to 4.5 and a strength of 22 to 25 g per tex. Indian cottons conforming to long and extra long staple group are either too fine or too weak. There is an urgent need to promote those cottons that could come closer in quality to the most sought after foreign cottons (Sreenivasan, 2001).

Earlier cotton industries were giving preference in the order of fibre length, fibre fineness, fibre maturity and at the last fibre strength. But changes in spinning technology have created a requirement for unique and greater fibre strength. Because of rigors in the ginning, opening, cleaning, carding, combing and drafting the important objective has been the improvement of the fibre strength (Zhang *et al.*, 2003). Because of modernization of textile industry through increased automation of spinning and weaving, there is demand for the change in the order of preference of fibre quality parameters for making cotton quality to be comparable and competitive with the man made fibres. There is an urgent need to develop cotton varieties or hybrids for high fibre strength (>25 g/tex) as the fibre strength in present day cultivars especially upland and *desi* -cotton is just any where between 15 to 22 g/tex.

Wild species of *Gossypium* are rich with rare desirable attributes that are not available in the germplasm of cultivated species. Hence, conservation of wild germplasm along with other genetic stocks forms an integral part of breeding strategy in cotton improvement. Hybridization between species is undertaken either for exploiting the hybrid vigour or to incorporate desirable gene or constellation of genes into cultivated species. Hence, introgression of character through interspecific hybridization is resorted to enrich the genetic heritage of the cultivated species. In addition to high yielding varieties and hybrids, the future demands for new plant types suited to varied agro-climatic regions, cotton for easy care properties with multi-adversity resistance *etc*. These could be achieved only by introgressive hybridization utilizing wild species, which are the rich sources for these characters (Vinitha and Phundhan Singh, 2004). The possibilities of evolving interspecific commercial hybrid between *G. hirsutum* and *G. barbadense* with extra long, fine, strong and silky fibre were indicated in early 1950s (Patel and Patel, 1959). The first of such interspecific hybrid between American and Egyptian cotton known as Varalaxmi was released in 1972 (Katarki, 1981). Katageri and Kadapa (1989) identified bollworm tolerant interspecific hybrids.

Diploid cottons belonging to *G. herbaceum* and *G. arboreum* were cultivated on 90 per cent of the total cotton cultivated area before independence. But today their cultivation is restricted to 27 per cent only (Singh and Narayanan, 1991). Herbaceum cottons have wider adaptability and higher degree of resistance to biotic (insect and disease) and abiotic (drought and windstorm) stresses (Patel and Mehta, 1989 and 1990). These *desi* cottons have deep root system and less vegetative growth which helps in withstanding drought conditions. Pubescent nature provides resistance to sucking pests and also have storm proof boll character which protects shedding of locules against strong winds after boll opening. Similarly arboreum cottons are usually coarse and short fibred, but due to their high moisture absorbancy, they have greater export value. As diploids are suitable only for producing 20 count yarn, the market value for these cottons is lesser than tetraploids. Development of diploid cotton suitable for spinning 40 count yarn will make them more remunerative for the diploid cotton growers. Balancing of characteristics like high yielding ability, superior fibre properties, abiotic and biotic stress tolerance could be possible by introgressing desirable characteristics between diploid and tetraploid cottons.

Wild species of cotton which form the bulk of the genus *Gossypium* are the source of genes for resistance to pests and diseases, with good fibre properties. As *G. tomentosum* is known to contribute high fibre strength, there is need to transfer this trait to cultivated species to meet textile needs. *G. anomalum* Wawra and Peyritsch lintless cotton is known for some superior fibre properties like high strength and fineness, transfer of these traits to the cultivated species is need of the hour.

For molecular tagging of fibre quality and other traits, there is a necessity of constructing molecular maps using suitable mapping population. It requires assessment of genetic diversity and generation of recombinant lines by interspecific hybridization and selection for further planned use. Morphological features are indicative of the genotype but are represented by only few loci because they are not large enough. Moreover, they can also be affected by environmental factors and cultural practices. To have an accurate and reliable estimate of genetic relationships and genetic diversity assessment molecular markers can be used as they provide an unlimited number of marker loci (Williams *et al.*, 1990).

With this in view, to improve the fibre properties of *G. herbaceum* var. DDhc-11, *G. anomalum* was chosen as donor parent in the present study with the following objectives.

1. To study the genetic introgression of morphological characters.
2. To study the genetic introgression of yield and yield contributing characters.
3. To study the genetic introgression of fibre properties.
4. To study the genetic diversity through molecular markers.

II. REVIEW OF LITERATURE

Wild species of *Gossypium* are rich with rare desirable attributes that are not available in the germplasm of cultivated species. Hence, conservation of wild germplasm along with other genetic stocks forms an integral part of a meaningful breeding strategy in cotton improvement. Hybridization between species is undertaken either for exploiting the hybrid vigour or to incorporate desirable gene or constellation of genes into cultivated species (Vinitha and Phundhan Singh, 2004).

The key factor for the success of plant breeding is availability of genetic variability for several characters which can be achieved through interspecific hybridization and mutations. The successful introgression of genes via interspecific hybridization has resulted only after extensive recombinations and selections (Stalker, 1980). Interspecific hybridization between two different species, but cultivated types having same ploidy and belonging to same genera is not a difficult task. Several workers have used distant hybridization for genetic improvement in cotton (Singh, 1998).

Interspecific hybridization in cotton between cultivated tetraploid species has been exploited in developing commercially popular cotton hybrids like Varalaxmi and Jayalaxmi (Katarki, 1971 and 1981). Similarly, interspecific hybrids between cultivated diploid species were carried to commercialize *desi* hybrids like DH7 and DH9 (Mehta *et al.*, 1985 and 1988). But it is not so easy to cross species belonging to different ploidy and between cultivated and wild species belonging to different genomic groups. The risk involved in this is due to the incompatibility at different stages of fertilization and zygote development (Stebbins, 1958).

Evolution of cultivated allopolyploids in different crops are first standard examples of cross between different diploid species. The genus, *Gossypium* comprises 50 species spreading over eight genomic groups (‘A’ through ‘G’ and ‘K’). Four species are cultivated and exploited commercially as fibre crop. 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 cultivated diploid (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. It indicates clearly that wild type contributed positively to fixable heterosis for yield and superior fibre quality. Many breeding stocks as well as varieties have been evolved so far through interspecific hybridization involving wild species. *Hirsutum*-Arboreum-Raimondi (HAR) and Arboreum-Thurberi-*Hirsutum* (ATH) lines were produced in 1940s. 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*. Development of cytoplasmic male sterile lines utilizing the cytoplasm of wild species is a great boon to seed industry producing commercial cotton hybrid seeds (Vinitha and Phundhan Singh, 2004).

Some of the selected genetic introgression studies carried out are discussed under various headings.

2.1 GENETIC INTROGRESSION STUDIES FOR MORPHOLOGICAL CHARACTERS

Amin (1940) made cross between the *G. herbaceum* x *G. barbadense* and observed resemblance of hybrids with their maternal parent for most of morphological characters.

Reduction in anthocyanin pigmentation on leaf was recorded by Amin (1940) in a cross between Red arboreum with American cotton and for most of morphological characters they resembled female parent.

Sreerangaswamy and Raman (1963) crossed *G. herbaceum* (cultivated) with *G. anomalum* (wild species). Hybrids were vigorous with broader leaves and size of petals exceeded the parents and the size of bracteole was similar to that of wild parent. The bolls resembled the wild parent in many aspects, such as pitted glands, shape of bolls and size of bracteoles.

Katarki (1971) carried out interspecific hybridization between *G. hirsutum* x *G. barbadense* and the F_1 was intermediate for plant height and leaf lobing and the characters transferred from male parent were petal spot and pubesense.

Thombre and Mehetre (1981) crossed *G. arboreum* male sterile with *G. anomalum* as male, the F_1 had boll shape and fibre colour of the male parent and leaf shape, bract shape and leaf hairiness of female parent.

Gill and Bajaj (1984) studied the morphology of hybrids between Asiatic diploid (*G. arboreum* and *G. herbaceum*) species and wild diploid (*G. stocksii* and *G. anomalum*) species. The hybrids were intermediate in leaf, flower and bract characters. In the cross between *G. arboreum* and *G. anomalum*, the hybrid was vigorous and produced large number of flowers compared to their parents as well as other hybrids. All the hybrids were perennial in nature.

Considering the limitations of interspecific transfer of tetraploid traits, Deshpande *et al.* (1992) tried autopolyploidization of diploid *G. arboreum* to obtain autotetraploid (2n=52, $4A_2$) type. The autotetraploid so obtained was further crossed with *G. hirsutum* genome to produce interspecific hybrid with 2n=4x=52, $2A_2AD_1$ genome. This had resulted in hastening the process of introgression unlike previous workers. The F_1 had 27.6 per cent pollen fertility and they observed that leaf shape, plant habit, petal spots, petal colour and pollen colour were similar to *G. arboreum* while, bud and flower size resembled that of tetraploid.

Among six upland cotton restorer lines reported by Davis (1993), line 4 showed a high proportion of introgression from *G. barbadense* but resembled upland cotton in many morphological characters.

Genetic diversity was studied at phenotypic level by Tatineni *et al.,* 1996 using stable and highly heritable morphological characters which easily distinguish *G. barbadense* and *G. hirsutum* and he noticed Several genotypes that were phenotypically distinct from typical *G. hirsutum* and *G. barbadense*.

All the F_1 generation plants from cross between *G. hirsutum* × *G. barbadense* dominated the *G. barbadense* for phenotype and frequency of *G. hirsutum* phenotype progressively increased from F_2 to F_5 generations with plant height, boll weight and earliness (Galanopoulou-Sendouca and Roupakias, 1999).

Brar *et al.* (1999) obtained F_1s from three interspecific crosses *viz*., *G. arboreum* cv. Lo.327 × *G. thurberi*, *G. arboreum* L-617 × *G. anomalum* and *G. herbaceum* cv. G. cot 11 × *G. anomalum* which were intermediate for most of the morphological characters and were sterile.

Kachapur (2001) evaluated the plants which are derived from cross between Jayadhar x BCS-23 for all the morphological characters of leaf, stem, bract, flower and boll and he noticed the intermediate nature of hybrids for most of morphological characters.

Crosses made between American and Asiatic cottons differing in ploidy level revealed that the cross recovery was 0.7 and 0.1 per cent when tetraploids and diploids were maternal parent respectively (Feng, 1935; Katageri *et al.,* 1999 and Rajashekhar *et al.,* 2003).

Manohar (2003) noticed fifteen different groups in a cross between DS-28 × SB(YF)-425 based on four distinguishable morphological characters namely petal spot, pubescence, branching habit and leaf lobing and all the groups except two were one or the other way was combination of *hirsutum* and *barbadense* type.

Soregaon (2004) reported in the cross between DS-28 × SB(YF)-425 eight different groups based on four distinguishable morphological characters namely petal spot, pubesense, branching habit and leaf lobing. He observed nearly 55 per cent of the plants falling under a group consisting two characters each from both the parent and none of the plants showed all four characters from any of the parent.

Veeresh Gowda *et al.* (2004) studied morphological characters of the cross between *G. herbaceum* var. DDhc-11 × *G. anomalum* in which he used 22 plants of F_2, 13 plants of BC_1 and 3 plants BC_2 generation and only distinguishable characters has been studied. Among the 8 plants of F_2, 7 plants of BC_1 and 2 plants of BC_2 were recorded as recombinants based on morphological characters.

2.2 GENETIC INTROGRESSION STUDIES FOR YIELD AND YIELD CONTRIBUTING CHARACTERS

The primary focus in the cotton products is usually higher yield rather than fibre quality. The high value per ha of cotton and the recent demands for high quality of fibres clearly justify the importance of hybrids and varieties, which are superior in yield.

Hutchinson *et al.* (1945) obtained vigourous types with larger bolls than sea island and also types whose seed showed an exceptionally good cover of lint in the second back-cross of *G. barbadense* x *G. raimondii* hexaploid with sea island cotton. Ganeshan (1946) observed that the progenies derived from cross between *G. hirsutum* x *G. raimondii* hexaploid with cultivated *G. hirsutum* cotton possessed fine and stronger lint and also high vegetative cover.

In a cross between cultivated (*G. herbaceum*) and wild species (*G. anomalum*), Sreerangaswamy and Raman (1963) harvested 68 bolls which yielded 178 seeds and the cross recovery ranged between 9.17 to 11.74 per cent. They also studied the morphology of the hybrids. Hybrids were vigorous with broader leaves. Size of petals exceeded than in parents and the size of bractiole was similar to that of wild parent. The bolls resembled the wild parent in many aspects, such as pitted glands, shape of bolls and size of bractioles.

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) F_l X 1627 A.L.F. Which possessed ginning out turn 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 gl_2 and gl_3 (both for glandlessness), or genes closely linked to these therefore appear to depress lint yield without affecting fibre properties.

Omel-chenko *et al.* (1979) made interspecific hybridization of varieties of *G. hirsutum* (AD_1) and *G. barbadense* (AD_2) with *G. tomentosum* (AD_3, resistant to sucking pests owing to its hairiness has strong fibre). They reported that 5.1-22.0 per cent of recombinants possessed traits absent in their parents and economically useful high yielding plants with strong fibre and resistance to sucking pests were obtained among these recombinants.

Arutyunova and Volkova (1987) reported in a cross between *G. hirsutum* and *G. stocksii*, the F_1 had low boll setting on selfing, but on crossing with *G. hirsutum* there was high boll set. Sherzhanova (1987) hybridized ecologically distant forms of cotton with *G. hirsutum i.e.*, Chindz and Tashkent-6 with Acala SJ-4 and Coker, to produce high yielding forms, resistant to *Verticilium dahliae*.

McCarty *et al.* (1995) reported that 16 accessions of *G. hirsutum* were crossed with Deltapine 16 and progeny with day neutral flowering habit were selected in the F_2 generation. These progenies were back crossed four times to their respective primitive accession parent. The resulting progenies (F_5, BC_1F_5, BC_2F_5, BC_3F_5 and BC_4F_5) were evaluated for yield and fibre traits for 3 years. Finally authors concluded that lint yield and lint percentage tended to decrease with successive back crosses; whereas, seed size and fibre strength tended to increase with additional back crosses.

Linted hybrid with several intermediate characters were recovered from cross between *G. herbaceum* var. DDhC-11 and lintless diploid *G. anomalum* by Soregaon (2004).

Studies carried out by Gururajan and Sundar (2004) revealed that the existence of significant positive association between yield per plant and seed index, boll number and boll weight, whereas 2.5% span length was negatively correlated with yield per plant. Path coefficient analysis revealed that the lint index, boll number and boll weight had highest direct positive effect on yield per plant and hence direct selection can be made for these traits.

Nirania *et al.* (2004) reported in a study that was undertaken to identify superior combining parents for yield and quality traits in a line × tester analysis found out that among male parents A-72-15 was the best general combiner for seed cotton yield followed by H777. Similarly, HP Acala for maturity coefficient and GS 9 for 2.5% span length were identified as best combiners. With regard to GOT, fibre fineness and bundle strength, male parents *viz.*, Tamcot SP37H, N183 and J2P7, respectively were identified as the best general combiners.

Iyanar *et al.* (2004) reported that the culture TCH 1649 has registered the highest yield of 3871 kg per ha which showed 13.1 per cent and 113.7 per cent increase over check varieties Surabhi (3422 kg/ha) and MCU 5 (1811 kg/ha), respectively. TCH 1648 and TCH 1649 have been identified as the best derivatives with improved yield, quality and biotic stress tolerance through introgressive breeding.

In the study conducted by Kajjidoni *et al.* (2004) reported that out of 12 different morphological, seed cotton yield and economic traits, bolls per plant and lint index exhibited highest direct effect in positive direction and ginning out turn in negative direction inspite of their significant association with seed cotton yield per plant at genotypic level in positive direction in both sets of hybrids.

Siva Prasad *et al.* (2004) using line × tester (7 lines and 6 testers) mating design in American cotton revealed that both additive and non-additive gene effects were important for days to 50 per cent flowering, seed index, ginning percentage, 2.5% span length, micronaire, maturity coefficient and seed cotton yield per plant. Non-additive gene action was observed for number of monopodia per plant, number of sympodia per plant, number of seeds per boll and lint index.

In a cross between *G. hirsutum* var. DS-28 × *G. barbadense* var. SB (YF) – 425 (Veeresh Gowda, 2004) reported that mean seed cotton yield, seed index, lint weight, ginning outturn and lint index of recombinant plants were superior over DS-28. In a cross between *G. herbaceum* var. Jayadhar × *G. barbadense* var. BCS-23, the same author reported that seed cotton yield, seed index, lint weight, ginning outturn and lint index are significantly higher than Jayadhar. In a cross between *G. arboreum* var. A82-1 × *G. barbadense* var. BCS-23, he also reported that seed cotton yield, seed index, lint weight, ginning outturn are significantly superior than A82-1. In the cross between *G. herbaceum* var. DDhc-11 × *G. anomalum* also he observed the same results.

2.3 INTROGRESSION STUDIES FOR FIBRE PROPERTIES

Competition from synthetic fibres and the need to improve fibre quality are two major economic forces driving the current global cotton market. Technological changes in the textile industry demands higher quality fibre particularly fibre strength, fineness and fibre length.

Investigation on the cross between *G.hirsutum* x *G. tomentosum* reported by Dark (1960) and through intensive selection over a considerable period by five back crosses, a genotype with long lint was derived.

To transfer fibre fineness from *G. anomalum*, Apparthurai *et al.* (1964), crossed cultivated coarse cotton species (*G. arboreum*) with *G. anomalum*. They obtained partially fertile F_1 plants which were back crossed with *G. arboreum* to get fine quality cotton strain in this species.

Katarki (1971) carried out 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).

From the studies of El-Hattab *et al.* (1974) in F_4-F_6 generations of the interspecific crosses *viz.*, Giza 67 (*G. barbadense*) x Deltapine 15 (*G. hirsutum*) and Giza 45 x Acala 4-42 (*G. hirsutum*) they noticed that earliness had significant negative correlation with fibre length except in the F_6 of Giza 67 x Deltapine 15 which had positive and highly significant correlation. They also reported that in two F_6 lines of Giza 67 x Deltapine 15, fibre length (1.18-1.19 inch) more than that of Giza 67 had been obtained.

Meyer (1974) transferred characters such as nectariless from *G. tomentosum*, fibrous root character from *G. sturti*, increased fibre strength from *G. thurberi*, boll worm 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.

The extra long staple breeding line Sealand 542 (GP150), a selection from the fourth back-cross generation of 'Sea Island' (*G. barbadense*) x 'Coker Wilds' (*G. hirsutum*, recurrent parent) had important genes for fibre strength (Culp and Harrell, 1980).

Introgressive gene transfer in *Gossypium* spp. was reviewed by Narayanan *et al.* (1984) and reported that, in India successful hybridization between *G. hirsutum* and *G. tomentosum* resulted Badnawar-1, B1007 and other CTI type cultivars and they concluded that transfer of desirable genes from wild relatives into cultivated background is an effective way of conservation and also preventing genetic erosion.

The nectariless trait 2 (conferred by the genes ne1 and ne 2 and contributing to the insect resistance) was introgressed into *G. hirsutum* germplasm from a wild Hawaiian species (*G. tomentosum*). The HVI fibre strength of MD51ne and Deltapine 50 had shown an average of 30.9 and 24 mN/tex, respectively in 29 comparisons (Meredith, 1993).

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.

Two sets of long linted, fine *G. arboreum* genotypes Dharwad Long Staple Arboreum Cotton (DLSA) and PA were developed by Kulkarni and Khadi (1998) through interspecific hybridization and selection. These were evaluated for yield and quality under rainfed conditions and they reported fibre length upto 30.37 mm with micronaire values between 4.0 and 5.0. The fibre strength was between 19 to 21 g/tex (1/8" gauge).

Recombinant *G.hirsutum* lines with superior fibre properties from *G. barbadense* were obtained by Manohar (2003) and they recorded 30-33 mm fibre length and fibre strength of 22-24 g/tex.

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).

Recombinants of segregating population (F_4) of cross between *G. hirsutum* var. DS-28 and *G. barbadense* var. SB (YF)-425 were evaluated by Soregaon (2004) and he noticed 23 recombinant plants with fibre length and strength more than 30 mm and 22 g/tex respectively. In the cross between Jayadhar x BCS 23, he noticed superiority in fibre length (24-26mm), fibre strength (20-23g/tex) than Jayadhar (20mm fibre length and 16-17g/tex fibre strength). He also observed recombinants from cross between A82-1 (*G. arboreum*) x BCS 23 (*G. barbadense*) which had fibre length of 24.9 mm (2.5% SL) and 25.9 g/tex fibre strength with 3.3 micronaire value as against 17.9 mm, 18.9 g/tex and 5.7 respectively of A82-1.

Veeresh Gowda (2004) reported that recombinant plants from cross between *G. hirsutum* cv. DS-28 × *G. barbadense* cv. SB (YF)-425 of F_6 generation recorded superior fibre length (32.5 mm) and fibre strength (27.2 g/tex) with higher per cent increase over DS-28 for seed cotton yield and yield contributing traits. Similarly, in the cross *G. herbaceum* cv. Jayadhar × *G. barbadense* cv. BCS-23, recombinants with fibre length (32.0 mm), fibre strength (27.4 g/tex) and micronaire value around 4.0 were selected from BC_2, BC_1, F_2 and F_3 populations, which were superior in terms of yield. Balanced recombinants having higher fibre length (23.8 mm), fibre strength (24.8 g/tex) and micronaire value (3.8) along with higher per cent increase in SCY and yield contributing traits were derived at BC_2 and BC_1F_2 generation of cross *G. arboreum* cv. A82-1 × *G. barbadense* cv. BCS-23. Similarly, plants with elevated fibre properties were selected from BC_1, BC_2 and F_2 of cross between *G. herbaceum* cv. DDhc-11 × *G. anomalum.* Recombinants with 27.0 mm fibre length and 23.0 g per tex fibre strength were isolated at F_4 from cross between *G. hirsutum* cv. Abhadita × (G. cot-11 × *G. tomentosum*).

2.4 STUDIES THROUGH MOLECULAR MARKERS

Morphological features are indicative of the genotype but are represented by only a few loci because they are not large enough. Moreover, they can also be affected by environmental factors and cultural practices. To have an accurate and reliable estimate of genetic relationships and genetic diversity assessment, there is a need of polymorhic molecular markers. Therefore, RAPD technique (Williams *et al.,* 1990) provides unlimited number of marker loci that can be used for genetic diversity and genetic recombination analysis. It was successfully applied to characterize germplasm lines developed through interspecific hybridization. RAPD markers have been used for the estimation of genetic similarities and cultivar analysis for introgressed genes through amplified genomic regions.

Limited interspecific introgressions have been achieved in the breeding of two primarily cultivated species of cotton *viz., G. hirsutum* and *G. barbadense* (Tatineni *et al.,* 1996). Genetic diversity was investigated at the DNA level using the RAPD and at the phenotypic level using stable and highly heritable morphological characters which easily distinguish *G. barbadense* and *G. hirsutum*. Eighty random decamer primers were used to amplify DNA *via* PCR and 135 RAPDs were generated. Dendrograms were generated and both procedures produced two clusters with one resembling *G. hirsutum* and the other *G. barbadense*. Several genotypes were identified that were genetically and phenotypically distinct from typical *G. hirsutum* and *G. barbadense*.

Iqbal *et al.* (1997) used RAPD analysis to evaluate the genetic diversity of elite commercial cotton varieties. Twenty-two varieties belonging to *G. hirsutum* and one belonging to *G. arboreum* were analysed with fifty random decamer primers using the polymerase chain reaction (PCR). Fourty nine primers detected polymorphism in all twenty three cotton varieties while, one produced monomorphic amplification profile. They reported that, a total of 349 bands were amplified, of which 89.1 per cent were polymorphic. The diploid cotton *G. arboreum* cv. Ravi was also distinct from rest of its tetraploid counterpart and showed only 55.7 per cent similarity.

Khan *et al.* (2000) carried out an experiment in *Gossypium* species to evaluate genetic diversity by RAPD using 45 random decamer primers. OPJ-17 produced the maximum number of fragments while, the minimum number of fragments were produced with primer OPA-08 and 99.8 per cent bands were polymorphic. Cluster analysis showed considerable disagreement among the D-genome (diploid new world species) and C-genome (diploid Australian species) species. Finally they concluded that, the interspecific genetic relationship of several species is related to their centre of origin. The results also revealed the genetic relationship of the species *G. hirsutum* with standard cultivated *G. barbadense*, *G. herbaceum* and *G. arboreum*.

Polymorphism and genetic similarity of five germplasm lines derived from [4 x (*G. arboreum* x *G. anomaium*)] x *G. hirsutum* and their parents were evaluated through RAPD markers generated by 22 random decamer primers. The result showed that there was abundant DNA polymorphism in three parents. Special DNA fragment of parents appeared in five germplasm lines. Lower similarity coefficients between different *Gossypium* species were related to their centre of origin and genome difference whereas, higher similarity coefficient between germplasm lines was due to back crossing by *G. hirsutum* generation to generation (Nie *et al.,* 2000).

Yadav *et al.* (2001) utilized the inbred lines, DS-28 (female) and SB(YF)-425 (male) and then they were screened with random primers for DNA polymorphism. Using pollen parent specific random primers, the genetic purity of DCH-32 hybrid seeds was determined by random amplified polymorphic DNA (RAPD) analysis. The results from RAPD for different hybrid seed lots of DCH-32 agreed well with the field based grow out test. It is suggested that RAPD method might be an alternative to the time consuming grow out test.

Rana and Bhat (2002) employed RAPD analysis in commercially released 18 cultivars of diploid cotton belonging to two species *i.e.*, *G. arboreum* and *G. herbaceum*. They selected 20 primers from 60 initially screened, generated 224 amplification products of which 127 amplicons were polymorphic. All selected primers produced polymorphic amplification products across genotypes. Cluster analysis showed clear-cut separation of the genotypes of the two species where, *G. herbaceum* cv. Jayadhar did not cluster with any other cultivar. *G. arboreum* cultivars formed three clusters and cluster II contained three out of four cultivars from the same place.

Fifteen genotypes belonging to *G. hirsutum* and one belonging to *G. arboreum* were analysed by Manickam *et al.* (2002) with 18 random decamer primers using the polymerase chain reaction (PCR). A total of 1275 bands were amplified; 62.7 per cent of which were polymorphic. First cluster was formed by all tetraploid genotypes and their hybrids and the second cluster was formed by the single diploid genotype (K10).

A haploid population from the cross of the two cultivated allotetraploid cottons, *G. hirsutum* and *G. barbadense* (cv. Hai 7124), was developed by Zhang *et al.* (2002). A molecular linkage map was constructed with 58 doubled and haploid plants. Among the total of 624 marker loci (510 SSRs and 114 RAPD), 489 loci were assembled into 43 linkage groups and covered 3314.5 centi-Morgans (cM).

Abdel Ghany *et al.* (2003) investigated the molecular nature of RAPD fragments in four *G. barbadense* cultivars. Five RAPD fragments, generated by improved RAPD-PCR technique representing polymorphic and nonpolymorphic bands were analyzed at the molecular level using DNA sequence analysis. Nonpolymorphic RAPD fragments showed homologies to previously characterized plant structural genes. Comparative nucleotide sequence analysis of two co-migrating nonpolymorphic fragments revealed that these two DNA sequences are highly similar to each other, indicating that similarity of fragment size is a good predicator of homology. Polymorphic RAPD fragments, on the other hand showed homologies to middle and high-repetitive DNA sequences. These results promote the initiative to integrate these RAPD markers in cotton breeding applications and DNA fingerprinting.

Genetic diversity in a set of 30 elite cotton germplasm lines was studied by Pawan kumar *et al.* (2003) which included 20 genotypes of *G. hirsutum*, seven genotypes of *G. arboreum,* one genotype each of *G. herbaceum*, *G. thurberi* and *G. klostzschianum*. For RAPD analysis they used 32 random primers in which only 25 showed amplification while rest failed to amplify. A total of 108 bands were amplified in 30 genotypes. They reported one primer, OLIGO 656 which amplified an 1100 bp *G. arboreum* specific band.

RAPD was used to evaluate genetic diversity among 7 diploid (*G. arboreum*) and fifteen tetraploid (*G. hirsutum*) cotton cultivars. RAPD markers were efficient and detected 88 per cent polymorphism. The diploid and tetraploid cultivars could be divided into separate cluster at 30 per cent similarity. RAPD revealed higher diversity among diploids than among tetraploids (Vafaie-Tabar *et al.,* 2003).

Zhang *et al.* (2003) used a *G. anomalum* introgression line 7235 characterized by good fibre quality properties to identify molecular markers linked to fibre strength QTLs. They used of F_2 and F_3 populations derived from a cross between 7235 and TM-1, a genetic standard of upland cotton. Nine molecular markers, three SSRs and six RAPDs linked to two QTLs for fibre strength has been identified. They reported a major QTL *i.e.,* QTL $_{FS1}$ found to be associated with eight markers and explained more than 30 per cent of the phenotypic variation.

Ashok Dongre *et al*. (2004) used forty five ISSR and forty RAPD primers for characterization of twenty five cotton gemplasms. Out of these, 19 ISSR and 21 RAPD primers are scorable and they generated ninety and one hundred and fifty markers respectively. Forty six markers from ISSR and seventy six markers from RAPD were scored as polymorphic. Dendrograms were developed for ISSR and RAPD analysis. Dendrogram of ISSR analysis showed three clusters while that of RAPD showed four clusters. They also used the six agronomic traits for charecterization of germplasm. Significant similarities were found in the clustering of ISSR and RAPD analysis. However correlations were not made between the clustering of ISSR, RAPD analysis and agronomic data analysis.

Genetic diversity analysis among recombinants from cross between Jayadhar x BCS 23 was carried out by Soregaon *et al.* (2004) using nine primers. On an average 78.26 per cent polymorphism was observed and the presence of higher genetic diversity was evidenced by higher range of genetic similarity indices.

Gomes *et al.* (2004) used RAPD markers to assess the genetic relatedness in seven *Gossypium* species which included *G. hirsutum,* CMS lines and cultivars, *G arboreum* CMS lines, cultivars and wild species, *G. raimondii, G. bickii, G. thurberi, G. captis-virdis* and *G. anomolum*. Out of forty five RAPD primers tested, twenty four primers yielded monomorphic amplified products and remaining twenty one primers amplified a total of one sixty eight fragments with an average of 9.8 fragments per primer. Out of eleven genotypes studied, *G. arboreum* (G 27) showed maximum number of DNA amplified fragments.

A genetic linkage map of tetraploid cotton was constructed using SSRs, RAPDs and SRAPs by Xianlong *et al* (2004). A total of three sixty eight SSR primer pairs, six hundred RAPD primers and two thirty eight SRAP primer combinations were used to screen polymorphism between parents and revealed seven hundred forty seven loci in total two hundred five SSRs, one hundred seven RAPDs and four hundred thirty seven SRAPs. Sixty-nine F_2 individuals from the interspecific cross of *Gossypium hirsutum* 'Handan 208' and *G. barbadense* 'Pima 90' were genotyped for the seven forty seven polymorphic markers

Yingzhi Lu *et al.* (2004) developed an upland cotton germplasm, NM 24016 derived from an interspecific cross between *G. hirsutum* and *G. barbadense* was used to develop two recombinant inbred line (RIL) populations. A total of approximately five hundred markers, consisting of SSRs, RAPDs, STSs, AFLPs, and SRAPs have been developed for the two mapping populations (TM-1 x NM24016 and 3-79 x NM24016). The STS markers were converted from their original RAPD markers. Markers associated with morphological traits, fibre quality and yield have been identified.

Hanchinal *et al.* (2004) used RAPD markers to distinguish and identify the widely adapted DCH-32 and recently released DHB-105, interspecific cotton hybrids and their parents. They reported that three polymorphic bands distinguished the DHB-105 hybrid and its parents and results help for precise identification and hybrid purity testing of the above interspecific hybrids.

Vamadevaiah *et al.* (2004) reported that OPC-17 amplified a 700 bp locus in DHH-11, which is male specific, they reported using this marker male and female can be differentiated.

Zahid Mohammed *et al.* (2004) used RAPD technique to investigate the true ancestor species of Gossypium on the basis of 69 polymorphisms. The similarity coefficients were calculated and observed maximum similarity of 72 per cent between *Gossypium hirsutum* and its race latifolium, whereas minimum similarity of 30 per cent was observed between *Gossypium thurberi* and *G. hirsutum*. They concluded that genome AD of *G. hirsutum* comprises of *G. herbaceum* (A genome) and *G. raimondi* (D genome).

III. MATERIAL AND METHODS

The present investigation on genetic introgression studies in interspecific crosses of *Gossypium* species was carried out during *kharif* 2004-05 at Agricultural Research Station, Dharwad Farm, University of Agricultural Sciences, Dharwad.

3.1 DESCRIPTION OF THE EXPERIMENTAL MATERIALS

In the present study, segregating population (F_2) derived from cross between *G. herbaceum* var. DDhc-11 × *G. anomalum* was used. Characteristics of species including their salient features are presented in Table 1 and 2.

3.2 EXPERIMENTAL SITE LOCATION AND WEATHER CONDITION

Field experiments were conducted at Agricultural Research Station, Dharwad which is situated in the northern transitional zone (Zone no. 8) of Karnataka state with a latitude 15°17' N, longitude of 76°46' E and at an altitude of 678 meter above the mean sea level.

Experiments were laid out in black cotton soil and plots were homogenous with respect to nutrient status. Weather data is presented in Appendix I. The average rainfall for the year 2004-05 was 748 mm, which was poor both in terms of total precipitation and distribution. However, four protective irrigations were given at critical crop growth stages to get good crop stand in order to realize its potential expression for fibre and other properties. All other agronomic managements were followed according to recommended package of practices for irrigated condition of the zone.

3.3 METHODOLOGY

3.3.1 Layout

Individual plant to row progenies were raised with spacing mentioned in Table 3.

3.3.2 Observations recorded

The selection criterion was mainly to recover *G. herbaceum* var. DDhc-11 type of plants with elevated fibre properties.

3.3.2.1 Halo length measurement

About 218 plants were subjected to measurement of the halo length. Three bolls each picked at top, bottom and middle were used for measurement of halo length. Nine halo readings from three bolls per plant were taken. Mean halo length was calculated for each plant.

3.3.2.2 Morphological characters

Individual plants from the F_2 generation were considered for detailed study on morphological characters. Following morphological characters were studied.

1. *Colour of petal :* The petal colour was recorded as yellow, cream and light yellow.
2. *Petal spot :* The petal spot colour is recorded as purple, red and purplish red.
3. *Anther colour :* The colour of anther is recorded as yellow and cream.
4. *Filament colour :* The filament colour was recorded as cream yellow and purple.
5. *Bract type :* The bract type was recorded as normal, frego and intermediate.

Table 1: Morphological and economic characters of the cotton genotypes used in the study

Sl. No.	Genotypes	Characters	Genome	Ploidy
1.	*G. herbaceum* L. cv. DDhc-11	Cultivated cotton, pubescent stem, leaves 3-5 lobed, pubescent when young and glabrous at older age. Bracts flare widely from bud, flower and boll. Yellow petal with a red spot. Smooth bolls, drought resistant, sucking pest tolerant, short staple and low yielding	A_1	2n=2x=26
2.	*G. anomalum* Wawra and Peyritsch	Wild cotton, stellate-pubescent stem; leaves are ash green, petiolate and deeply five lobed (ovate), one to three inconspicuous foliar nectaries beneath on principal veins; petals pale yellow with a purple spot at base, sparsely and minutely gland dotted; seeds densely pubescent, the fibres brownish appressed and has good strength and fineness; resistance to leaf curl, boll worms, jassids, mites and nearly immune to black arm diseases; cytoplasmic male sterility.	B_1	2n=2x=26

Table 2: Distinguishable characters of *G. herbaceum* var. DDhc-11, *G. anomalum* and their F_1

Sl. No.	Character	Female (DDhc-11)	Male (*G. anomalum*)	F_1
1.	Leaf colour	Parrot green	Ash green	Ash green
2.	Leaf hairiness	Moderately hairy	Densely hairy	Densely hairy
3.	Petal colour	Yellow	Cream	Light yellow
4.	Petal spot	Red	Purple	Purplish red
5.	Anthers	Yellow	Cream	Yellow
6.	Filament	Cream yellow	Purple	Cream yellow
7.	Bract type	Normal	Frego	Intermediate
8.	Boll shape	Round	Tapered	Oblong
9.	Fibre colour	Milky white	lintless	Dull white
10.	Lint character	Linted	lintless	Linted

Table 3: Particulars of interspecific F_2 experimental material in cotton

Cross	**Population size**	*Generation*	**Date of sowing**	**Spacing (cm)**
G. herbaceum var. DDhc-11 (2x) × *G. anomalum* (2x)	218	F_2	25th June	90 × 20

3.3.2.3 Yield and yield contributing characters

The yield and yield contributing characters for all the 218 plants were recorded. Twenty six plants were selected based on halo length and lint availability and were subjected to paired 't' test.

a. Boll number : The total number of bolls of a plant.

b. Per boll weight (g) : Total kapas weight per boll

c. Seed cotton yield (g/plant) : The total quantity of seed cotton harvested from individual plant was taken as seed cotton yield per plant.

d. Seed yield (g) : Total seed weight of a plant.

e. Lint yield (g/plant) : The seed weight was substracted from seed cotton yield to calculate lint yield.

f. Seed index (g) : Weight of 100 seed was taken as seed index.

g. Ginning out turn (%) : Ginning out turn is the ratio of weight of lint to that of seed cotton, expressed in percentage as given below.

3.3.2.4 Fibre properties

Based on the availability of sufficient quantity of lint, 22 plants were subjected for detailed investigation for fibre properties. Following characters were analyzed under HVI (High Volume Instrument) at Central Institute for Research on Cotton technology (CIRCOT), Regional quality evaluation unit situated at ARS, Dharwad Farm.

a. 2.5% span length (mm) : 2.5% span length refers to the distance from the clamp on a fibre beared to a point upto which only 2.5 per cent of the fibres extended.

b. Fibre strength (g/tex) : It is the force required to break a fibre of unit linear density. Higher the force required, higher the strength of fibre.

c. Mironaire index (µg/inch) : It is average weight per unit length of fibre. It is used in determining the fibre fineness.

d. Fibre uniformity ratio (%) : It is the ratio between 50 per cent span length and the 2.5 per cent span length and it indicates the uniformity of fibre length.

e. Fibre maturity : It is an index of the extent of fibre development. The maturity depends mainly on the degree of secondary thickening of a fibre. The ratio of lumen and cell wall and less than one in case of mature fibres, 1-2 for half mature fibres and more than two for immature fibres.

f. Elongation percentage : A measure of per cent increase in jaw separation under load at the time of fibre rupture.

3.4 STUDIES THROUGH RAPD MARKERS

The F_2 population along with their parents and F_1 was subjected for RAPD analysis.

Genetic diversity analysis

Nine plants were selected with respective fibre strength values ranging from 17 to 25 g per tex. The individual plants DNA was analysed with RAPD primers.

Identification of molecular marker linked to fibre strength

The data of fibre strength was available for 22 plants. So these 22 plants were selected and made into nine groups representing the fibre strength values from 17 to 25 g per tex and these were screened using random decamer primers.

3.4.1 DNA extraction

The DNA was extracted from the recombinant lines and their parental genotypes by following CTAB extraction method (Saghai-Maroof *et al.*, 1984) with required modifications.

1. One or two fresh young leaves (not fully expanded and less than a week old) from shoot apex was harvested and the surface was cleaned with wet paper towel. The sample was ground to fine powder using liquid nitrogen with prechilled pestle and mortar.
2. The powder was transferred into a sterile 2.0 ml eppendorf tube. Then added 1 ml of extraction buffer (0.35 M sorbitol, 100 µM Tris and 5 µM EDTA). Before using, 0.4 g/100 ml of sodium bisulphite, 1 g/100 ml of PVP and 9 g/100 ml of glucose was added in to the extraction buffer.
3. The solution was vertexed for 30-40 seconds, centrifuged at 3000 rpm for 10 min.
4. Supernatant was discarded and to this 600 µl lysis buffer [1M Tris-HCl buffer pH8, 0.5 M ethylene diamine tetra acetic acid (EDTA) pH8, 5M NaCl and 2 per cent (w/v) hexadecyltrimethylammonium bromide (CTAB)] and 50 µl sarcosyl (5%) was added.
5. The solution was vortexed for 30-40 seconds.
6. The contents were incubated at 65°C in a water bath for 60 minutes with 2-3 times occasional shaking during incubation.
7. The tubes were kept for 10 min to allow them to return to room temperature.
8. 500 µl of chloroform and Iso amyl alcohol (CIA) solution prepared in a ratio of 24:1, was added to the tubes.
9. The solution was vortexed for 40-60 seconds and centrifuged at 8000 rpm for 10 minutes.
10. The aqueous supernatant solution was pipetted out and transferred to a fresh tube for DNA isolation. CIA extraction step was repeated.
11. Nucleic acids were precipitated by adding equal volume of ice cold isopropanol to the aqueous phase.
12. The solution was left for over night preservation at freezing temperature to enhance precipitation.
13. Solution was centrifuged at 8000 rpm for 5-10 minutes.
14. Supernatant was discarded (keeping pellet as it was).
15. The DNA pellet was washed with 70 per cent alcohol (400 µl) for 30 minutes and centrifuged at 8000 rpm for 10 minutes, pellet was recovered.
16. The tubes were allowed to drain and dried at room temperature for 2-3 hours
17. Then DNA was suspended in 500 to 700 µl of high salt TE (Tris EDTA) buffer (10 mM Tris HCl + 1mM EDTA + 2M NaCl, pH 7.5) in sterile eppendorf tube and kept for 45 minutes at room temperature.
18. When DNA was fully dissolved, 5 µl of RNase (10 mg/ml) was added and incubated at 37°C for one hour or at room temperature overnight.
19. An equal volume of cold phenol : chloroform : iso-amylalcohol (25:24:1 v/v/v) was added and mixed well. The solution was centrifuged for five minutes at 14,000 rpm.
20. Top layer was pipetted in to another eppendorf tube and an equal volume of chloroform : iso-amylalcohol (24:1 v/v) solution was added and centrifuged for five minutes at 14,000 rpm.

21. The top layer was pippetted in to new epperndorf tube and 0.5 volume of 7.5 mM ammonium acetate solution and two volumes of absolute ethanol was added, mixed thoroughly and centrifuged at 14,000 rpm for 5 minutes.

22. DNA pellet was removed and washed twice with 250 μl of 70% ethanol. The supernatant solution was dispensed and the pellet was air dried for 10 minutes.

23. The DNA was dissolved in 200 to 500 μl of TE (Tris-EDTA) buffer (10 mM Tris, 1 mM EDTA pH8).

3.4.2 DNA quantity and quality estimation

The concentration of DNA was assessed spectrophotometrically and also by gel electrophoresis using 0.8 per cent agarose with known concentration of uncut DNA.

In spectrophotometric analysis, 5 μl of DNA sample diluted with TE buffer and volume made upto 3000 μl with TE buffer and was subjected to spectrophotometer readings at absorbance of 230 nm, 260 nm and 280 nm. A good DNA preparation generally exhibits the following spectral properties.

$A_{230} < 0.10$, $A_{230}/A_{260} < 0.45$, $A_{280}/A_{260} < 0.55$, or $A_{260}/A_{280} > 1.80$

DNA concentration was calculated using O.D. at 260 nm with following formula.

Concentration of DNA (μg/ml) = O.D. at 260 x 50

To test the quality of DNA, samples were run on 0.80 per cent agarose gel in 1x TAE (Tris Acetic acid EDTA) buffer and stained with ethidium bromide and checked for contamination by RNA (which usually runs ahead) and the DNA was evaluated by comparing it with a standard undigested DNA sample.

3.4.3 Requirements for polymerase chain reaction

Template DNA: Purified genomic DNA (30 ng) of recombinants and their parents were used as template DNA per reaction.

Random primers: Commercial kits ‘A’, ‘B’, ‘C’, ‘P’, ‘Y’ and ‘Z’ of random decamer primers obtained from Operon Technologies Inc. Alamedas, USA were used.

In order to findout polymorphic primers for *G. herbaceum* var. DDhc-11 and *G. anomalum*, 100 random decamer primers *viz*., OPA 1 to 20 (10 primers), OPB series (10 primers), OPC series (20 primers), OPP series (20 primers), OPY series (20 primers) and OPZ series (20 primers) were used to screen two parents. As many as 21 primers which showed polymorphism were selected (Table 4). These primers were selected and were also used for linking a marker for fibre strength in the segregating population.

dNTPs: Individual dNTPs, dATP, dGTP, dCTP and dTTP obtained from M/S Bangalore Genei. Pvt. Ltd., Bangalore were used.

Taq DNA polymerase

Taq DNA polymerase (3 units per μl) and 10x *Taq* buffer were obtained from M/S Bangalore Genei. Pvt. Ltd., Bangalore.

Thermal cycler

Primus 96 plus supplied by MWG AG Biotech, Auzinger Strasse TA, Ebersberg, Germany was used for cyclic amplification of DNA.

3.4.4 Amplification reaction mixture

Amplification reaction mixture was prepared in 0.2 ml thin walled PCR tubes containing following components in a total volume of 25 μl.

Table 4: List of random decamer primers used for genetic diversity studies in cotton

Sl. No.	Primer	Sequence (5' – 3')
1.	OPA-2	TGCCGAGCTG
2.	OPA-9	GGGTAACGCC
3.	OPB-01	GTTTCGCTCC
4.	OPB-11	GTAGACCCGT
5.	OPC-1	TTCGAGCCAG
6.	OPC-9	CTCACCGTCC
7.	OPC-11	AAAGCTGCGG
8.	OPC-12	TGTCATCCCC
9.	OPC-20	ACTTCGCCAC
10.	OPP-6	GTGGGCTGAC
11.	OPP-7	GTCCATGCCA
12.	OPP-9	GTGGTCCGCA
13.	OPP-12	AAGGGCGAGT
14.	OPP-13	GGAGTGCCTG
15.	OPY-9	AGCAGCGCAC
16.	OPY-11	AGACGATGGG
17.	OPY-14	GGTCGATCTG
18.	OPY-18	GTGGAGTCAG
19.	OPY-20	AGCCGTGGAA
20.	OPZ-13	GACTAAGCCC
21.	OPZ-19	GTGCGAGCAA

Sl. No.	Components	Quantity (µl/Reaction)
1.	10x assay buffer with 15 mM $MgCl_2$	2.5
2.	$MgCl_2$	1.0
3.	DNTPs mix (2.5 mM each)	1.0
4.	Primer (5 pM/µl)	1.0
5.	Template (15 ng/µl)	2.0
6.	Sterile distilled water	17.16
7.	*Taq* DNA polymerase (3U/µl)	0.34

Except template, the master mix was distributed to PCR tubes (23 µl/tube) and later 2 µl of template DNA from the respective genotypes was added making the final volume of 25 µl.

3.4.5 The thermoprofile for PCR

The PCR amplification for RAPD analysis was performed according to Williams *et al.* (1990) with certain modifications. The amplification conditions were as follows.

Sl. No.	Step	Temperature (°C)	Duration (min)	Number of cycles
1.	Denaturation	95	5	1
2.	Denaturation	94	1	45
3.	Annealing	36	1	
4.	Extension	72	2	
5.	Final extension	72	8	1
6.	Dump	4	-	-

After the completion of the PCR, the products were stored at 4°C until the gel electrophoresis was done.

3.4.6 Separation of amplified products by agarose gel electrophoresis

The amplified products from each tube along with 2 µl of loading dye (bromophenol blue) were separated on 1.2 per cent agarose gel at 70 volts (<5 volts per cm of gel) using 1x TAE buffer of pH 8.0 containing ethidium bromide. Lambda DNA-EcoRI/Hind III double digest was used as DNA molecular weight marker. The gels were photographed using gel documentation system (Hero lab, E.A.S.Y. 440K).

3.4.7 Scoring the amplified fragments

The amplified fragmetns were scored as '1' for the presence and '0' for the absence of a band generating the 0 and 1 matrix and per cent polymorphism was calculated by using the following formula.

$$\text{Per cent polymorphism} = \frac{\text{Number of polymorphic bands}}{\text{Total no. of bands}} \times 100$$

3.5 STATISTICAL ANALYSIS

3.5.1 Statistical analysis

Qualitative morphological traits were recorded on all the 218 plants and they were subjected to chi-square test for fitting appropriate segregation ratios. χ^2 test was carried out using formula;

$$\chi 2 = \sum_{i=1}^{k} (O_i - E_i)^2 / E_i$$

Where,

O_i = Observed frequency of i^{th} class

E_i = Expected frequency of i^{th} class

χ^2 calculated value was compared with χ^2 table value at k – 1 degree of freedom. If χ^2 calculated value is greater than χ^2 table value, then it is said to be significant.

Yield characters for all the 218 plants were taken. Out of 218 plants, 26 plants were selected based on halo length and lint availability and these were subjected to paired 't' test. Like-wise based on fibre quality data availability, 22 plants were analysed using paired 't' test. Paired 't' test was carried out using MSTAT C software package.

3.5.2 Analysis of the profile of the amplified fragments

Pairwise genetic similarities (S_{ij}) between genotypes were estimated by DICE similarity coefficient. Clustering was done using the symmetric matrix of similarity coefficient and cluster obtained based on unweighted pair group arithmetic mean (UPGMA) using SHAN module of NTSYS-PC version 2.0 (Rohlf, 1998).

IV. EXPERIMENTAL RESULTS

The experimental results obtained in the present investigation on genetic introgression in interspecific cross of *Gossypium* species are presented under the following headings.

4.1 GENETIC INTROGRESSION STUDIES FOR MORPHOLOGICAL CHARACTERS

Five characters *viz*., petal colour, petal spot colour, anther colour, filament colour and bract type were recorded for 218 F_2 plants (Appendix II and Plate 1).

Fifty eight plants showed yellow petal similar to *G. herbaceum*, 43 plants showed cream petal, whereas 117 plants showed light yellow petal. As regards petal spot is concerned, 62 plants were having red petal spot similar to *G. herbaceum*, 51 plants were having purple petal spot similar to *G. anomalum*, whereas 105 plants were having purplish red petal spot.

One hundred and fifty two plants showed yellow anthers similar to *G. herbaceum* whereas, only 66 plants showed cream anthers similar to *G. anomalum*. In respect of filament colour, 172 plants showed cream yellow filament similar to *G. herbaceum* whereas only 46 plants showed purple filament colour and lastly for bract type, 68 plants showed normal bract type similar to *G. herbaceum*, 48 plants showed frego bracts similar to *G. anomalum*, whereas 102 plants showed bract type which was intermediate between both the parents. The segregation ratios in which these fit are given in Table 5.

Morphology of high lint yielding plants

The 26 plants which were considered for evaluation of all fibre quality tests based on lint availability were put into seven groups (Table 6) based on character recombination for petal spot, anther colour, filament colour and bract type.

Out of seven groups, group one possessed all the four characters resembling female parent DDhc-11. Plants in group number two possessed red petal spot, yellow anthers, and normal bract similar to DDhc-11 and purple filament which is similar to *G. anomalum*.

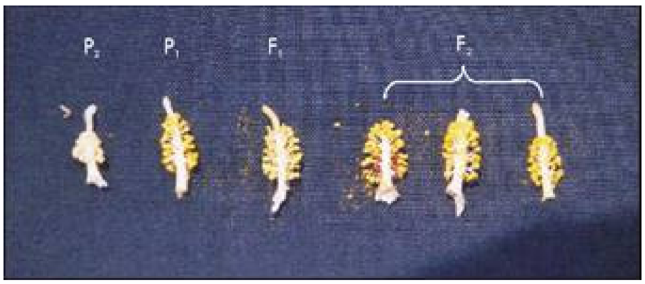
Plants in group number three possessed yellow anthers and normal bract like DDhc-11, but purple petal spot and purple filament similar to that of *G. anomalum*.

Plants belonging to group four possessed yellow anthers, cream yellow filament and normal bract like DDhc-11 and purple petal spot as that of male parent (*G. anomalum*). Plants belonging to group five possessed yellow anthers, cream yellow filaments and normal bract as that of DDhc-11 but purplish red petal spot which is intermediate between both the parents.

Plants belonging to group six possessed yellow anthers and normal bract similar to female parent, purple filament similar to male parent but possessed purplish red petal spot which is intermediate between both the parents and lastly the plants belonging to group seven possessed cream anthers, purple filaments and frego bracts similar to male parent and red petal spot similar to female parent (DDhc-11).

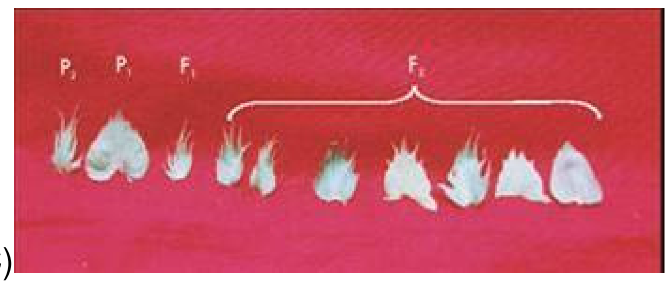
The recombinant plants were further grouped according to different proportions of characters from both the parents as shown in Table 7. Five plants were seen with 4H:0A (4 characters from *G. herbaceum* var. DDhc-11 and 0 characters from *G. anomalum*) combination accounts 19.23 per cent, eleven plants were seen with 3H:1A combination accounting 42.3 per cent, 2 plants with 2H:2A combination (7.69%), 2 plants with 1H:3A combination (7.6%), 4 plants were seen with recombinant and 3 *G. herbaceum* characters combination accounting 15.3 per cent whereas, two plants were seen with 1I:3H:1A combination accounting 7.69 per cent.

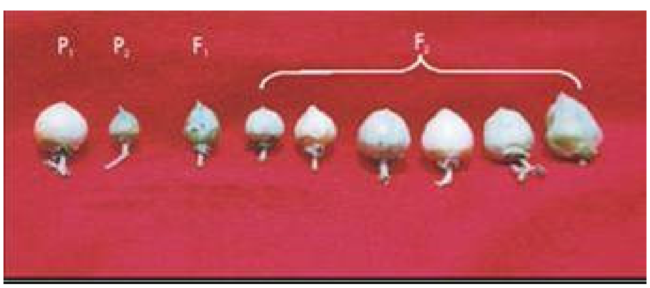
(A)



(B)

(C)





(D)

(E)

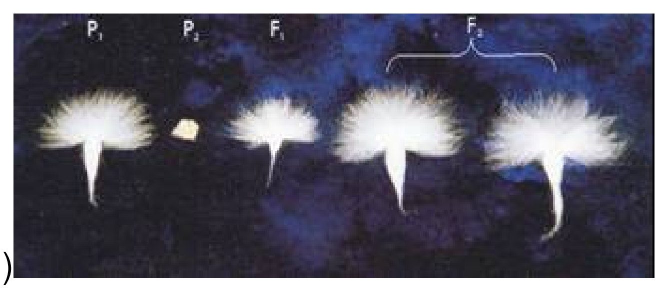


Plate 1: Genetic variability in F_2 for (A) Petal colour (B) Anther and filament colour (C) Bracttype (D) Boll shape (E) Halo length

4.2 GENETIC INTROGRESSION STUDIES FOR YIELD AND YIELD CONTRIBUTING CHARACTERS

Yield and yield contributing characters were recorded on all the 218 plants. The data of 218 plants were given in Appendix III. As many as 26 plants were selected based on halo length and sufficient lint availability and were considered for fibre quality analysis. They were subjected to 't' test for all the characters. The results of these 26 plants are presented in Table 8.

4.2.1 Number of bolls per plant

The mean boll number was 73.12, which was significantly superior over the mean boll number of DDhc-11 (49.81). The highest boll number was recorded by plant number 45 (237), followed by plant number 26 (159). The lowest boll number was recorded by 195 (10).

4.2.2 Seed cotton yield (g/plant)

The mean seed cotton yield of F_2 plants was 95.12 g, which was superior over the mean seed cotton yield of DDhc-11 (89.19 g) but it was not significant. The highest seed cotton yield was recorded by plant number 45 (269.0 g), followed by plant number 129 (194.0 g). The lowest seed cotton yield was recorded by 23rd plant (3.0 g) (Fig. 1).

4.2.3 Boll weight (g)

The mean boll weight of F_2 plants was 1.35 g, which was inferior to the mean boll weight of DDhc-11 (2.01 g). The highest boll weight was recorded by plant number 195 (2.3 g) followed by plant number 158 (1.91 g). The lowest boll weight was recorded by plant number 79 (0.67 g).

4.2.4 Seed index (g)

The mean seed index of F_2 plants (7.15 g) which was superior over DDhc-11 (7.05 g) but it was not significant. The highest seed index was recorded by two plants viz., 138 and 74 (7.6 g), followed by plants 3 and 133 (7.5 g), whereas the lowest seed index was recorded by plant number 182 (6.4 g).

4.2.5 Ginning out turn (%)

The mean ginning out turn (GOT) of the F_2 plants (30.04%) was significantly inferior to that of DDhc-11 (32.02%). The highest GOT was recorded by plant number 158 (34.62%) followed by plant number 74, 78 and 138 (34.0%), whereas the lowest GOT was recorded by plant number 182 (21.88%).

4.2.6 Lint index

The mean lint index of the F_2 plants (3.12) was inferior to DDhc-11 (3.25). The highest lint index was recorded by plant numbers 74 and 138 (3.92), whereas the lowest lint index was recorded by 194 (1.59).

4.3 GENETIC INTROGRESSION FOR FIBRE PROPERTIES

4.3.1 Halo length

The halo lengths of 218 F2 plants ranged between 14.44 to 26.33 mm with a mean value of 19.58 mm. The data is presented in Appendix IV. Maximum number of plants (115) recorded halo length in the range of 19.1 to 22.0 mm category while, only three plants recorded more than 25.1 mm halo length. The other classes viz., 13.1 to 16.0 mm, 16.1 to 19.0 mm and 22.1 to 25.0 contained 5, 78 and 17 plants, respectively (Fig. 2).

Table 5: Morphological characters of F_2 plants in an interspecific cross of cotton

Character	*No. of plants*	**Fits in segregation ratio**	**Chi square value**
Petal colour	Yellow (58) Light yellow (117) Cream (43)	1:2:1	3.237
Petal spot colour	Red (62) Purplish red (105) Purple (51)	1:2:1	1.402
Anther colour	Yellow (152) Cream (66)	3:1	3.228
Filament colour	Cream yellow (172) Purple (46)	3:1	1.766
Bract type	Normal (68) Intermediate (102) Frego (48)	1:2:1	4.568

Table 6: Frequency of qualitative morphological characters of selected F_2 plants in an interspecific cross of cotton

Group	**Phenotype**		**Number of plants**
1.	Petal spot Anther colour Filament colour Bract type	Red Yellow Cream yellow Normal	5
2.	Petal spot Anther colour Filament colour Bract type	Red Yellow **Purple** Normal	4
3.	Petal spot Anther colour Filament colour Bract type	**Purple** Yellow **Purple** Normal	2
4.	Petal spot Anther colour Filament colour Bract type	**Purple** Yellow Cream yellow Normal	7
5.	Petal spot Anther colour Filament colour Bract type	***Purplish red*** Yellow Cream Normal	4
6.	Petal spot Anther colour Filament colour Bract type	***Purplish red*** Yellow **Purple** Normal	2
7.	Petal spot Anther colour Filament colour Bract type	Red Cream **Purple** **Frego**	2
	Total		**26**

Regular : *G. herbaceum* var. DDhc-11 character
Bold : *G. anomalum* character
Italic bold : Intermediate character

Table 7: Frequency of selected plants falling under different groups representing combinations based on four qualitative morphological features in F_2 of an interspecific cross of cotton

Combination of characters	4H:0A	3H:1A	2H:2A	1H:3A	1I:3H	1I:2H:1A
Number of plants	5	11	2	2	4	2
Per cent of plants	19.2	42.3	7.69	7.69	15.30	7.69

H – Indicates characteristics of *G. herbaceum* var. DDhc-11 (red petal spot, yellow anthers, cream yellow filament and normal bract type)

A – Indicates characteristics of *G. anomalum* (purple petal spot, cream yellow anthers, purple filaments and frego bracts)

I – Intermediate characters (purplish red petal spot)

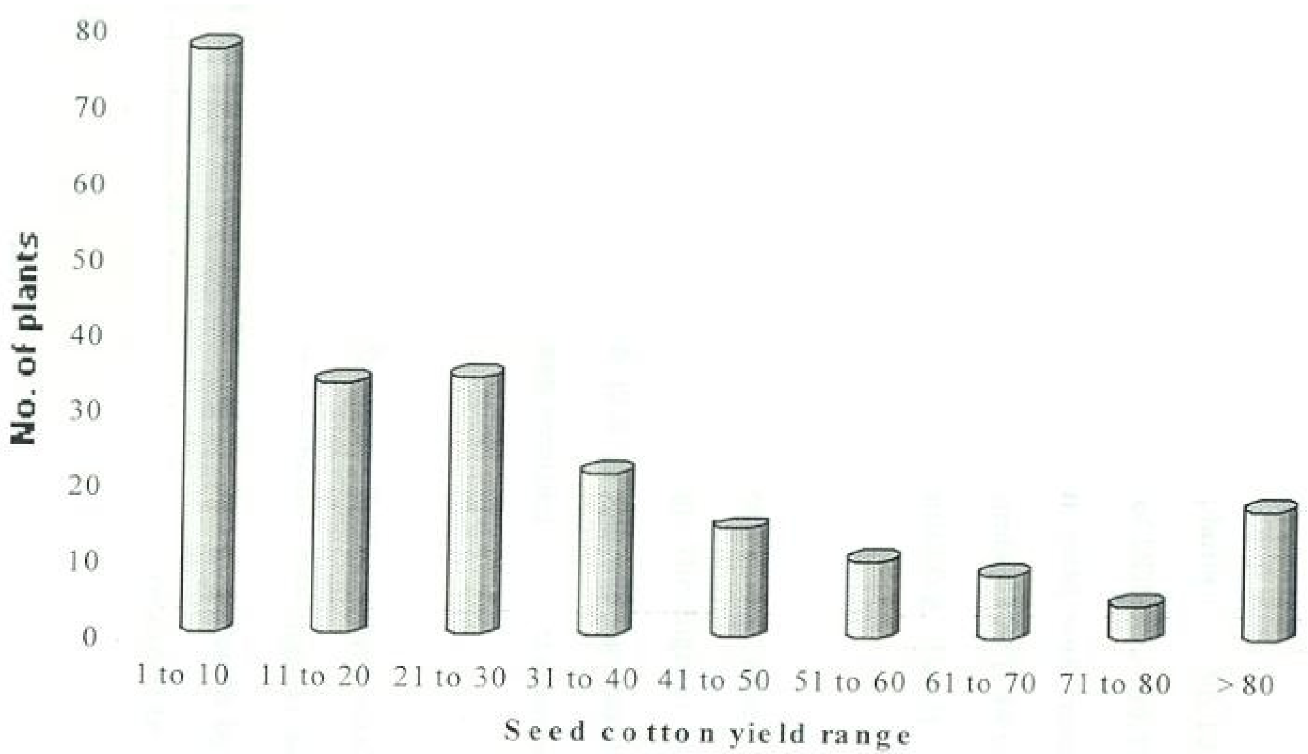


Fig. 1: Frequency distribution of F_2 segregants of an interspecific cotton cross for seed cotton yield

Table 8: ***Per se*** **performance of F_2 segregants of an interspecific cross of cotton for yield and yield contributing characters**

Sl. No.	Pedigree	No. of bolls per plant		Seed cotton yield (g)		Per boll weight (g)		Seed index (g)		Ginning outturn (%)		Lint index	
		DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2
1.	3	48	106	86	88	2.4	0.83	7.1	7.5	32.3	27.9	3.2	2.9
2.	16	52	98	94	132	1.9	1.34	6.9	7.2	34.3	24.5	3.4	2.3
3.	17	56	87	82	92	2.4	1.05	7.2	7.2	29.5	33.3	2.9	3.6
4.	23	47	13	84	3	1.8	1.00	6.9	6.5	31.5	30.0	3.1	2.8
5.	26	49	159	89	188	2.1	1.18	7.2	7.4	33.2	29.2	3.5	3.0
6.	32	51	22	86	23	2.1	1.04	7.2	7.1	29.9	31.3	3.4	3.2
7.	34	54	80	91	84	2.2	1.05	7.2	7.4	30.5	33.3	3.1	3.7
8.	42	48	41	94	59	1.8	1.43	7.1	7.3	32.4	32.6	3.2	3.5
9.	45	49	237	86	269	1.8	1.13	6.8	7.3	33.5	28.6	3.2	2.9
10.	65	52	46	95	58	2.1	1.26	7.1	7.1	29.8	33.3	3.4	3.6
11.	74	46	64	89	67	1.8	1.04	7.2	7.6	33.6	34.0	3.1	3.9
12.	78	54	50	92	58	1.8	1.16	7.1	7.4	32.1	34.0	3.2	3.8
13.	79	47	85	95	57	2.3	0.67	7.2	7.1	30.4	31.3	3.4	3.2
14.	129	49	115	88	194	2.2	1.68	6.9	7.0	33.5	30.6	3.6	3.1
15.	130	52	66	94	99	2.1	1.50	7.0	7.3	33.4	33.3	3.2	3.7
16.	131	45	118	87	184	1.8	1.55	6.9	7.2	32.5	29.5	3.1	3.0
17.	133	46	78	86	107	1.9	1.37	7.2	7.5	33.4	31.3	3.2	3.4
18.	136	45	54	84	82	2.2	1.51	7.1	7.1	32.5	30.5	3.5	3.1
19.	138	58	44	95	69	1.9	1.56	6.9	7.6	31.6	34.0	3.4	3.9
20.	143	43	64	86	102	1.7	1.59	6.8	7.2	32.5	31.6	3.5	3.3
21.	155	48	53	89	71	2.2	1.33	7.1	7.2	31.2	31.1	3.1	3.3
22.	156	50	37	86	39	2.2	1.05	7.1	6.8	33.2	18.9	3.2	1.6
23.	158	49	69	88	132	2.1	1.91	6.9	7.2	32.6	34.6	3.2	3.8
24.	182	51	17	90	31	1.8	1.82	7.2	6.4	31.5	21.9	3.1	1.8
25.	195	52	10	92	23	1.8	2.30	6.8	6.5	30.5	25.0	3.2	2.2
26.	203	54	88	91	162	1.9	1.84	7.2	6.9	31.2	25.3	3.1	2.3
	Mean	49.81	73.12*	89.19	95.12	2.01	1.35	7.05	7.15	32.02*	30.0	3.25	3.1

	S.D.	3.63	48.64	3.84	62.61	0.21	0.37	0.15	0.32	1.34	4.0	0.17	0.6
	t' test	0.24		0.47		0.51		1.73		2.32		1.48	
	S.D.#	49.2		63.2		0.45		0.30		0.44		0.71	
	SEm$\pm$	9.67		12.42		0.09		0.06		0.87		0.14	

* - Significant at 0.05 level of probability # - Combine SD for DDhc-11 and F_2

4.3.2 Fibre properties

The data of fibre properties of 22 plants for which sufficient amount of lint was available is presented in Table 9. Data was subjected to paired 't' test. The results are presented below.

4.3.2.1 Fibre length (2.5 per cent SL, mm)

The mean fibre length of F_2 plants (23.51 mm) was superior over the mean fibre length of DDhc-11 (23.11 mm), but it was non significant. The longest fibre length was recorded by plant number 129 (25.5 mm), followed by plant number 203 (25.4 mm) and plant number 156 (25.2 mm) whereas, the shortest fibre length was recorded by plant number 34 (20.3 mm).

4.3.2.2 Fibre strength (g/tex)

The mean fibre strength of the F_2 plants (21.01 g/tex) was significantly superior than the mean fibre strength of female parent *i.e.*, DDhc-11 (19.16 g/tex). The highest fibre strength was recorded by plant number 203 (25.2 g/tex), followed by plant number 3 (24.0 g/tex) whereas the lowest fibre strength was recorded by plant number 16 (17.6 g/tex).

4.3.2.3 Fibre elongation

The mean fibre elongation of the F_2 plants (7.0%) was found significantly inferior to DDhc-11 (7.31%). The highest fibre elongation (7.5%) was recorded by plant number 26, followed by two plants 136, 130 (7.4%) whereas, the lowest fibre elongation (6.5%) was recorded by two plants 3 and 155.

4.3.2.4 Fibre fineness (micronaire value)

The mean micronaire value of the F_2 plants (4.16 μg/inch) was significantly lower than DDhc-11 (5.09 μg/inch). The lowest micronaire value was observed in plant number 203 (2.4 μg/inch), whereas highest micronaire value was observed in two plants *viz*., 16 and 130 (5.7 μg/inch).

4.3.2.5 Uniformity ratio (%)

The mean uniformity ratio of the F_2 plants (50.68%) was superior to the mean uniformity ratio of DDhc-11 (50.5%) but it was not significant. The highest uniformity ratio was recorded by two plants *viz*., 34 and 17 (54%), whereas lowest uniformity ratio was recorded for four plants 203, 78, 74 and 42 (49%).

4.3.2.6 Fibre maturity ratio

Highest maturity ratio (0.88) was recorded by plant number 130 followed by plant number 16 (0.87) whereas, the lowest maturity ratio was recorded by plant number 17 (0.63). The mean maturity ratio of recombinant plants (0.75) was superior over mean maturity ratio of DDhc-11 (0.72), but it was not significant.

4.4 MOLECULAR MARKERS ANALYSIS

4.4.1 Identification of polymorphism between parents

Out of 100 primers screened, only 21 primers were found to be polymorphic between parents and also they were repeatable. Out of these polymorphic primers OPP-12 was found to be the most informative recording 80 per cent of polymorphism. The same 80 per cent of polymorphism was also recorded for other primers like OPC-12 and OPZ-19, but the number of bands were low. The primer OPZ-13 was found to be the least polymorphic rather it was monomorphic (Appendix V).

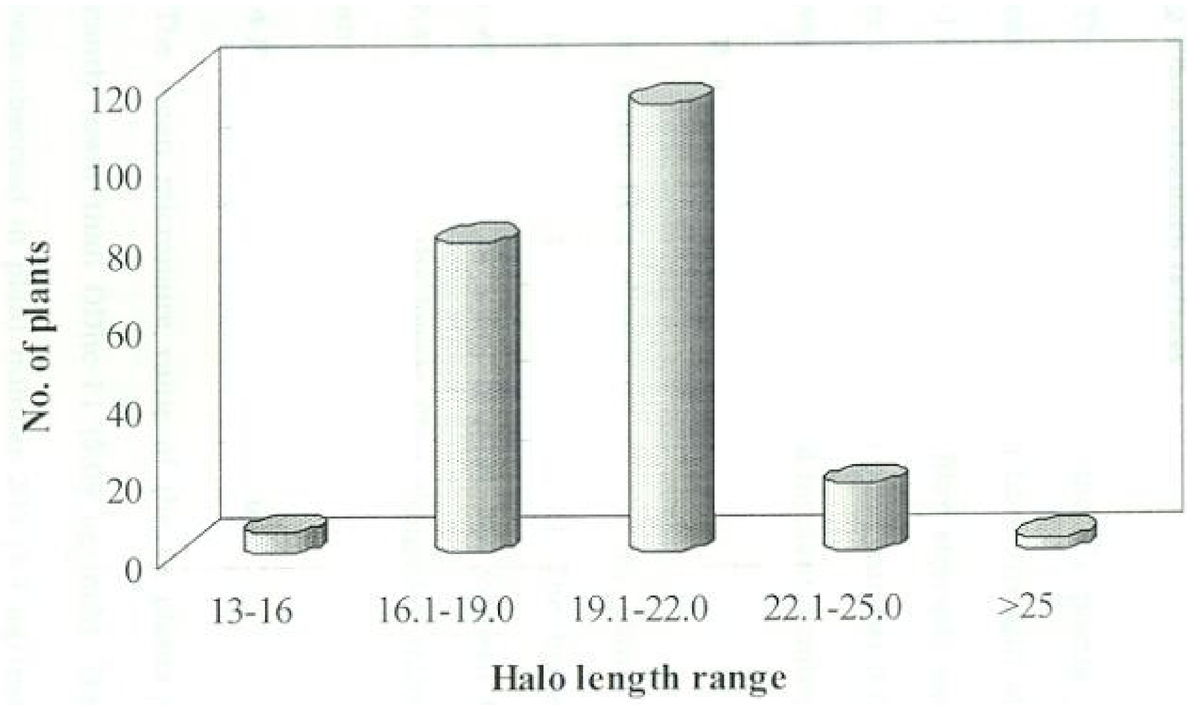


Fig. 2. Frequency distribution of F_2 segregants for halo length in an interspecific cross of cotton

Table 9: Fibre properties of F_2 segregants in an interspecific cross of cotton

Sl. No.	Pedigree	Fiber length (2.5%SL) (mm)		Uniformity ratio (%)		Fibre fineness (Micronaire)		Maturity ratio		Fibre strength (g/tex)		Elongation (%)	
		DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2	DDhc-11	F_2
1.	3	23.4	23.4	49.0	51.0	5.2	3.2	0.8	0.6	18.4	24.0	7.1	6.5
2.	16	23.1	20.6	50.0	51.0	5.5	5.7	0.7	0.9	18.7	17.6	7.3	7.1
3.	17	23.2	21.9	52.0	54.0	5.4	3.0	0.8	0.6	19.1	23.0	7.4	6.7
4.	26	23.3	20.9	52.0	52.0	5.3	4.4	0.7	0.8	19.5	19.2	7.1	7.5
5.	34	23.3	20.3	49.0	54.0	5.1	4.4	0.7	0.8	19.6	19.1	7.5	7.1
6.	42	23.4	25.0	51.0	49.0	5.3	3.9	0.7	0.7	19.5	20.4	7.0	6.9
7.	45	23.3	22.3	50.0	51.0	4.9	4.4	0.7	0.7	19.1	21.0	7.5	6.8
8.	65	23.2	23.2	49.0	50.0	5.1	3.2	0.8	0.7	18.8	22.1	7.3	7.0
9.	74	23.8	24.7	53.0	49.0	4.9	3.4	0.7	0.7	18.6	21.9	7.1	6.9
10.	78	23.8	22.2	49.0	49.0	5.2	4.0	0.7	0.7	19.3	19.4	7.4	7.2
11.	79	23.2	23.0	49.0	50.0	5.3	3.9	0.7	0.7	19.5	21.4	7.6	6.6
12.	129	23.1	25.5	52.0	50.0	5.3	4.2	0.8	0.8	19.7	21.8	7.0	7.1
13.	130	22.9	24.3	51.0	50.0	5.4	5.7	0.8	0.9	19.6	19.2	7.3	7.4
14.	131	22.8	24.5	51.0	51.0	5.0	4.4	0.8	0.8	19.8	21.1	7.2	7.0
15.	133	22.7	24.8	50.0	51.0	4.8	4.4	0.7	0.8	19.8	21.6	7.4	6.8
16.	136	22.8	23.9	50.0	51.0	4.6	4.2	0.7	0.8	19.6	20.7	7.6	7.4
17.	138	22.7	24.0	51.0	50.0	5.1	3.6	0.7	0.7	18.5	21.1	7.4	7.0
18.	143	22.6	24.0	51.0	50.0	4.9	4.7	0.6	0.8	19.0	20.3	7.0	7.2
19.	155	23.8	25.1	50.0	52.0	4.8	4.4	0.8	0.8	18.9	22.8	7.0	6.5
20.	156	22.8	25.2	49.0	50.0	5.0	4.6	0.7	0.8	18.5	20.9	7.4	7.3
21.	158	22.7	22.7	51.0	51.0	5.0	5.4	0.7	0.9	18.6	18.5	7.6	7.3
22.	203	22.5	25.4	53.0	49.0	4.9	2.4	0.7	0.6	19.5	25.2	7.6	6.6
	Mean	23.11	23.50	50.55	50.68	5.09**	4.16	0.72	0.75	19.16	21.01**	7.31**	7.00
	S.D	0.39	1.59	1.30	1.39	0.23	0.83	0.05	0.07	0.47	1.81	0.22	0.30
	T test	1.91		0.09		4.21		0.03		4.59		4.80	

	S.D.#	0.87		2.01		0.98		0.43		1.89		0.35	
	SEm±	0.18		0.43		0.20		0.09		0.40		0.07	

** - Significant at 0.01 level of probability # - Combine SD for DDhc-11 and F_2

Table 10: RAPD analysis for genetic diversity study in an interspecific F_2 population of cotton

Sl. No.	*Primers*	**Total No. of bands**	**No. of polymorphic bands**	**Per cent polymorphism**
1.	OPC-12	4	2	50.00
2.	OPB-11	8	5	62.50
3.	OPY-18	6	4	66.60
4.	OPP-6	7	4	57.10
5.	OPP-13	4	2	50.00
6.	OPY-11	7	5	71.40
7.	OPC-11	3	3	100.00
8.	OPB-01	5	3	60.00
9.	OPY-14	3	2	66.60
10.	OPA-2	6	4	66.60
11.	OPP-12	7	7	100.00
12.	OPZ-19	5	2	40.00
	Total	**65**	**43**	**66.10**
	No of bands per primer	**5.4**	**3.58**	

Table 11: Per cent contribution from male and female parents to F_2 based on molecular bands in cotton

Pedigree	**No. of specific bands from DDhc-11 (35)**	**Per cent specific bands from DDhc-11**	**No. of specific bands from *G. anomalum* (16)**	**Per cent specific bands from *G. anomalum***	**Monomorphic bands**	**Total No. of bands**
16	32	91.4	3	18.7	14	49
158	28	80.0	4	25.0	14	46
26	28	80.0	7	43.7	14	49
143	26	74.0	7	43.7	14	47
45	21	60.0	9	56.2	14	44
65	30	85.7	5	31.2	14	49
17	22	62.8	11	68.7	14	47
3	26	74.2	8	50.0	14	48
203	28	80.0	7	43.7	14	49
Average	26.7	76.4	6.7	42.3	14	47.5

4.4.2 Genetic diversity

Nine plants of cross between DDhc-11 × *G. anomalum* along with their parents were used for genetic diversity study. The nine genotypes were selected based on their fibre strength (17 to 25 g/tex). These nine genotypes were screened using 21 polymorphic primers. Out of these 21 primers only 12 primers showed amplification in all F_2 plants. They had shown two to seven polymorphic bands. Of the 65 amplified products from twelve primers, only 43 were polymorphic (66.1%). Total number of amplicons for each primer ranged from 3 for OPC-11 and OPC-14 to seven for OPP-12, OPY-11 and OPP-6 with an average of 5.4 amplicons per primer. Out of 5.4 amplicons, 3.58 amplicons per primer were polymorphic. The polymorphism per primer ranged from 50 (OPC-12, OPP-13) to 100 per cent (OPP-12) (Plate 2). The band profiles obtained with 12 primers are summarized in Table 11. Plant number 16 recorded highest contribution (91.4%) of specific bands from female parent followed by 80 per cent by plant numbers, 158, 203 and 26. Plant number 17 shown highest contribution (68.7%) from male parent (Table 12).

Cluster analysis and Dice similarity coefficients were carried out using NTSYS analysis package. The pair-wise similarity coefficient values for nine plants were calculated and are presented in Table 13. Overall similarity indices ranged from 60 to 82 per cent. The dendrogram constructed from the pooled data (Fig. 3) revealed two distinct clusters one in which only *G. anomalum* was present and in another, all the F_2 plants along with DDhc-11 were present. *G. anomalum* clustered distinctly from rest of the genotypes at 60 per cent similarity coefficient. The highest similarity coefficient was noticed between plant 1 and DDhc-11 with 82 per cent similarity coefficient. The F_2 plants were again subdivided into two clusters in one cluster plant 16, 65, 203 along with DDhc-11 were present in the other cluster remaining genotypes 158, 26, 143, 45 and 17 were present. The cluster analysis and similarity coefficient reveals that the genotypes selected were more closer towards the female parent DDhc-11.

4.4.3 Identification of marker linked to fibre strength

Twenty two plants with respective fibre strength values were made into nine groups representing fibre strength from 17.0 to 25.2 g per tex. These nine groups along with their parents and F_1 were screened with 21 polymorphic primers. A 300 bp fragment amplified from OPZ-19 had been noticed only in groups having fibre strength 23.0, 24.0, 25.0 g per tex along with P_2 and F_1 (Plate 2).

Table 12: Similarity matrix based on RAPD profile analysis for fibre strength in cotton

	DDhc-11	**1 (16)**	**2 (158)**	**3 (26)**	**4 (143)**	**5 (45)**	**6 (65)**	**7 (17)**	**8 (3)**	**9 (203)**	***G. anomalum***
DDhc-11	1.000										
1 (16)	0.824	1.000									
2 (158)	0.675	0.742	1.000								
3 (26)	0.674	0.739	0.767	1.000							
4 (143)	0.659	0.659	0.696	0.722	1.000						
5 (45)	0.608	0.635	0.697	0.638	0.677	1.000					
6 (65)	0.698	0.717	0.575	0.658	0.611	0.638	1.000				
7 (17)	0.721	0.674	0.685	0.711	0.667	0.696	0.605	1.000			
8 (3)	0.667	0.711	0.761	0.676	0.771	0.627	0.568	0.649	1.000		
9 (203)	0.683	0.682	0.551	0.583	0.500	0.492	0.611	0.528	0.514	1.000	
G. anomalum	0.506	0.624	0.568	0.675	0.603	0.571	0.675	0.675	0.533	0.521	1.000

Numbers within the parentheses indicate plant numbers

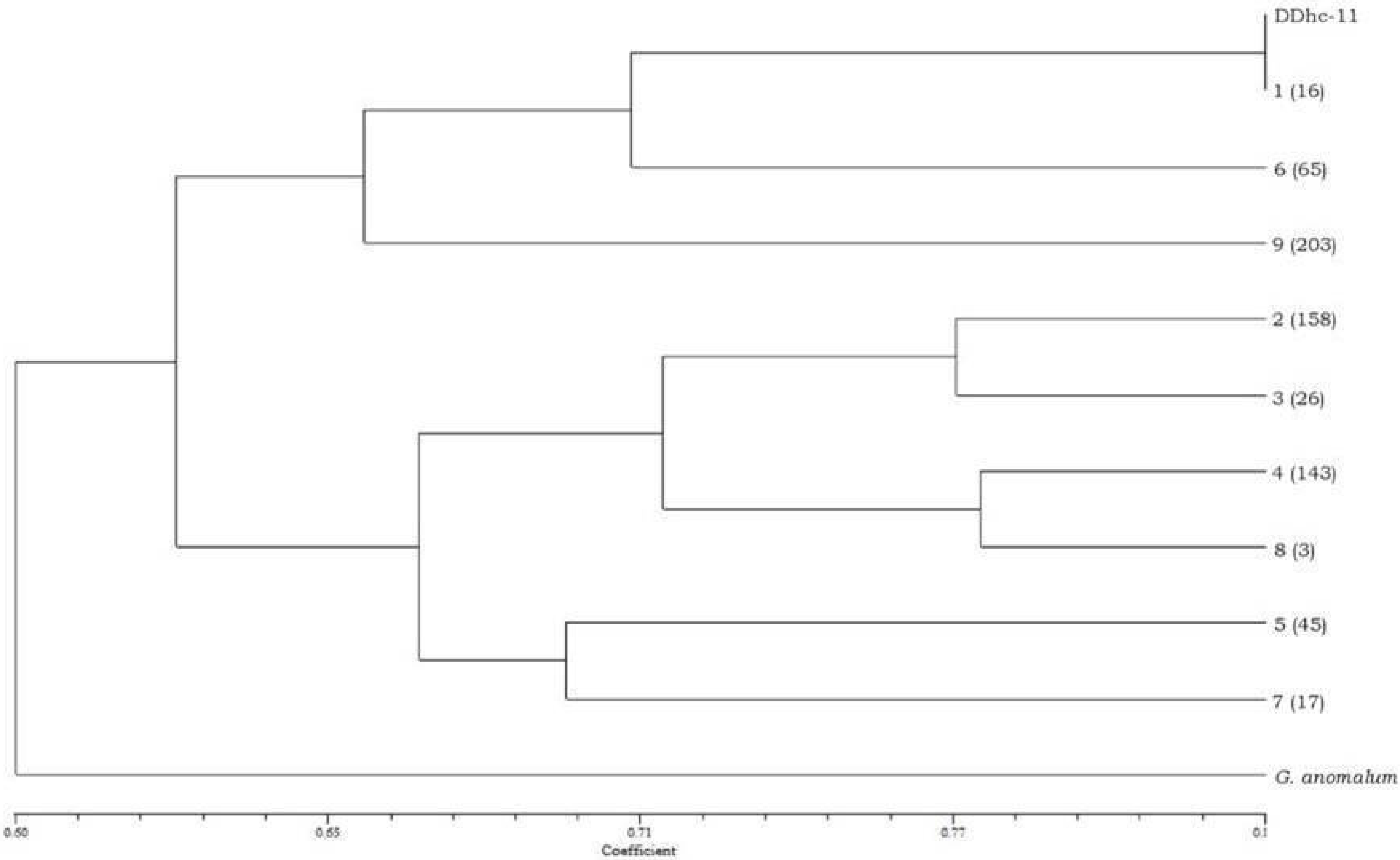
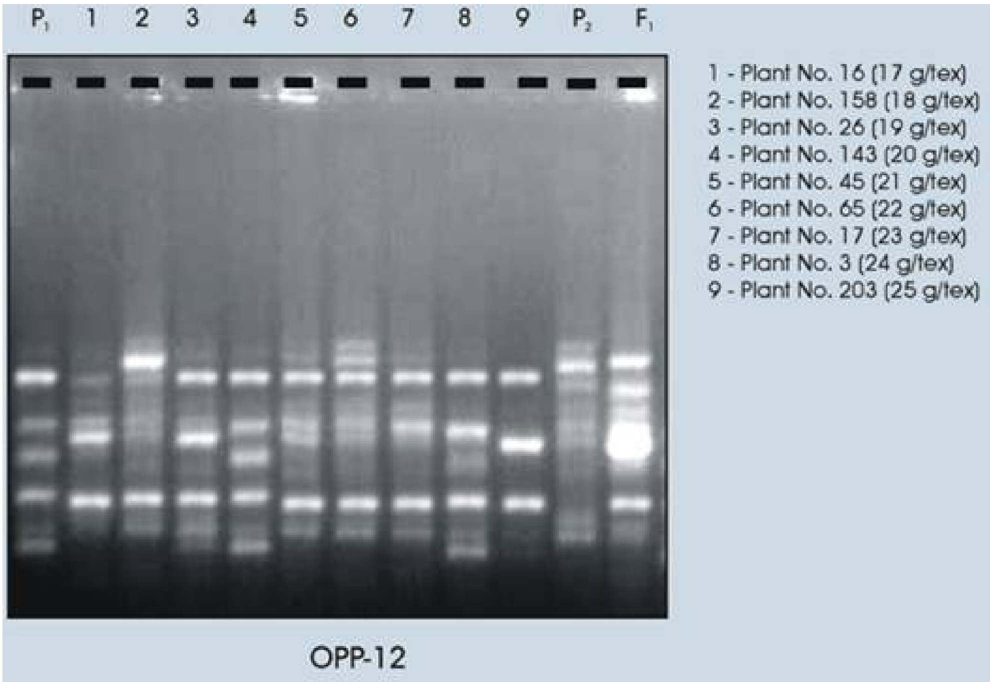


Fig. 3: Dendrogram generated from pooled data of RAPD profile using UPGMA analysis in an interspecific F_2 population



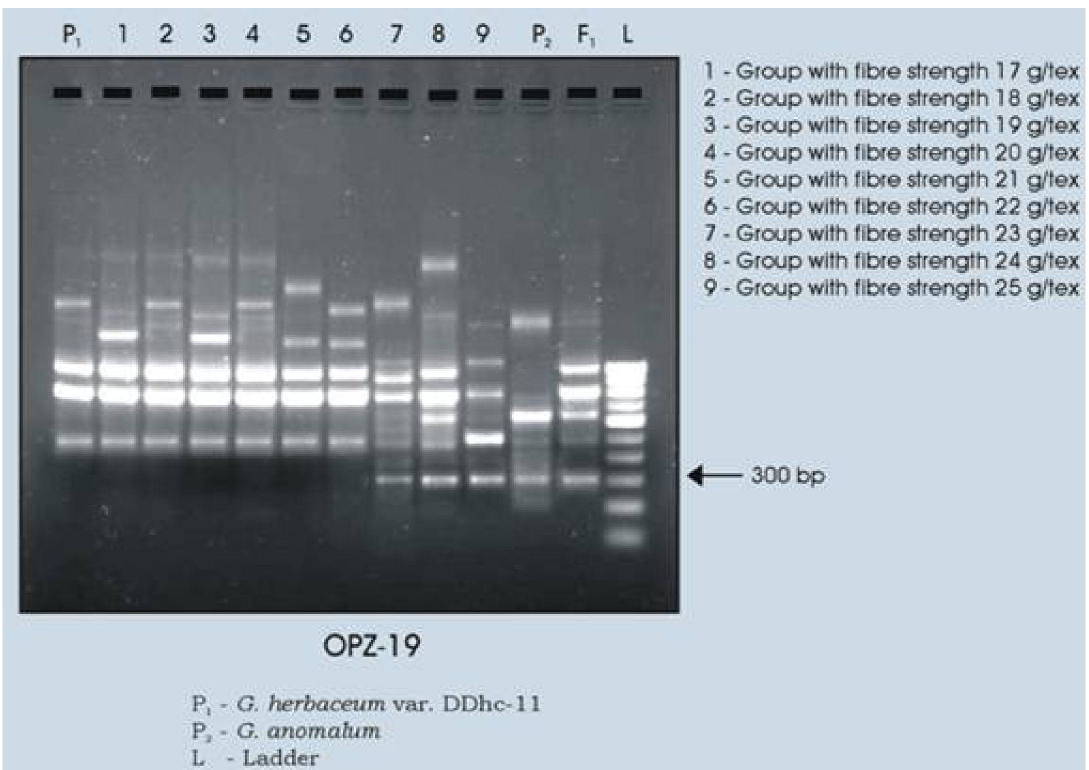


Plate 2: RAPD pattern in F_2 selected based on fibre strength

V. DISCUSSION

Wild species of *Gossypium* are rich with rare desirable attributes that are not available in the germplasm of cultivated species. Hence, conservation of wild germplasm along with other genetic stocks forms an integral part of a breeding programme in cotton improvement. Hybridization between species is undertaken either for exploiting the hybrid vigour or to incorporate desirable gene or a constellation of genes from wild into cultivated species. Hence, introgression of characters through interspecific hybridization is expected to enrich genetic heritage of the cultivated species with genes for more desirable economic characters. Intervarietal or intraspecific crosses are generally successful in the sense that they satisfy the conditions of maximum fitness due to which such hybrids are viable and fertile, hence equally favoured under both nature and domestication. In contrast, wide crosses suffer either from non-viability or sterility or from both. As a consequence, such distant hybrids or wide crosses are promptly eliminated by the forces of natural selection and so their perpetuation is discontinued. However, the skillful hand of man has made it possible to purposefully exploit many advantages of such remote hybridization in plant breeding.

Besides productivity, the future demands may be for new plant types suited to varied agro-climatic regions, cotton for easy care properties, varieties with multi-adversity resistance etc. These can be achieved by introgressive hybridization utilizing the wild species, which are the rich sources of these characters. Though, introgressive hybridization is time consuming, it is worth pursuing since there is ample scope for introducing rare genes (that are hithers to not available in cultivated gene pool) into the cultivated species to evolve superior varieties.

Introgression of wild germplasm into the cultivated cotton is difficult because of hybrid breakdown among interspecific hybrids reduced chiasma frequencies in many of the interspecific hybrids especially when the species involved in the hybridization are different at genome and ploidy level. There is also risk of introduction of undesirable genes along with the useful genes. The problems of interspecific incompatibility come into way of securing hybrids of some cross combinations, which could be overcome by utilizing embryo culture technique. Hybrid sterility is another hurdle, which is mainly due to difference in genome and ploidy level of species involved in the crosses. This could be overcome by synthesizing amphiploids. For transferring the desired characters, the amphiploid has to be backcrossed to the cultivated species repeatedly to obtain cytologically stable genotype with desired character. In this process, there is a risk of losing valuable genes present in the alien chromosomes, which usually tends to get eliminated. It takes several generations to obtain a genotype with introgressed characters. Use of molecular markers in selecting the plants with desired genes will greatly help in hastening this process.

It is generally difficult, though not impossible to achieve requisite success in interspecific hybridization largely because of well developed strategies to overcome interspecific barriers. The extent of crop improvement by interspecific hybridization can be assessed by the success stories in sugarcane, Triticum, Nicotiana and cotton. In cotton, there are 50 different species including four cultivated types. Interspecific hybridization has been extensively used in cotton to combine desirable characteristics, especially between species of same ploidy level. For the first time, commercial exploitation of interspecific hybridization was done in India by Katarki (1971) resulting in development of interspecific hybrid Varalaxmi. In this, recombination of high seed cotton yield, characteristic of *G. hirsutum* species and superior fibre quality characteristics of *G. barbadense* species was achieved. Since then, India has been earning foreign exchange with the export of long staple cotton obtained from many interspecific hybrids. Several desirable lines were derived from interspecific hybrids in cotton (Hutchinson, 1951; Knight, 1956; Pandya and Patel, 1956; Kadapa and Ganga Prasad, 1965; Kataraki, 1971, 1981; Katageri and Kadapa, 1989; Khadi, 1996 and Katageri *et al.*, 2003 and 2004).

Several studies indicated occurrence of haploids in different interspecific crosses (Skovsted, 1935; Kimber and Riley, 1963 and Lee, 1970). Thombre and Mehetre (1979) reported haploids from an interspecific cross between *G. hirsutum* var. Laxmi × *G. barbadense* var. SB-289E. Stephens (1949) reported that the degree of interspecific introgression in certain *Gossypium* hybrid population was often restricted by genetic breakdown in F_2 and subsequent generations. Interspecific F_1 hybrids between *G.*

barbadense and *G. hirsutum* are often vigourous and fertile, while selective elimination of certain genotypes and aberrant segregation occurs in later generation of selfing or backcrossing. Interspecific progenies frequently revert to phenotypes approaching either of the parents. According to Richmond (1951, eventhough large spectrum of variability can be released in progenies of interspecific cotton hybrids majority of the recombinants are ill balanced. Genetic differentiation of two genomes and small scale structural differences are considered to be limiting factors in isolating balanced recombinants. However, with all these associated problems of interspecific hybridization, some workers have achieved noticeable success. Harland (1936) indicated the possibility of developing interspecific types with desired features of *G. barbadense* and *G. hirsutum* species. Rajarathinam and Nadarajan (1993), Rajarathinam *et al.* (1993); Murthy *et al.* (1995a), Murthy *et al.* (1995b) and Kumar *et al.* (2000) reported that there is maximum contribution of one or more yield components towards genetic divergence.

As a result of advances in the spinning technology of present day textile industries, higher cotton fibre quality, especially strength is required. Development of genetic resources is a continuous process which helps to fulfill the changing demands/needs of consumers. The fibre properties of the present day varieties and hybrids are not able to meet the requirements of textile industry. To meet the present day demands of consumers, supply of high quality raw cotton is essential. The presence of mismatch between required fibre qualities and available fibre properties in the commercial varieties and hybrids necessitate the need for research on elevation of fibre properties of these varieties or hybrids. The present study was part of the research, which is going on in enhancement of fibre properties of commercial varieties at ARS, Dharwad and results of this study are discussed under the following headings.

5.1 GENETIC INTROGRESSION OF MORPHOLOGICAL CHARACTERS

Desi cottons are well adapted to wide range of climatic conditions. They are tolerant to both sucking pests and bollworms but are poor yielders as compared to hirsutum cotton varieties and hybrids, which are highly susceptible to pests and diseases. The farmers may prefer to grow desi cottons owing to low input cost required for their management and therefore there is need to develop high yielding varieties of desi cottons coupled with desirable fibre properties.

In the present study, it was aimed to recover the *G. herbaceum* var. DDhc-11 type of plants with elevated fibre properties using lintless wild relative *G. anomalum* from DDhc-11 × *G. anomalum* cross.

The possibility of deriving female parent type plants with elevated fibre properties has been shown in some other studies such as successful development of varieties like MCU-2 and MCU-3 of interspecific origin in Madras. The former is the ruling commercial variety grown in Cambodia Tract in Tamil Nadu during summer, while the latter derived from the combination of upland sea Morocco crosses, has shown its adaptability to Cambodia tract during winter by virtue of its vigour and tolerance to stem weevil and jassids (Santhanam and Krishnamurthy, 1961). Stable and near homozygous progenies were derived from interspecific hybrids that exhibited a wide range of combinations of parental morphological characteristics (Carnell and Davis, 1993).

In the present study, all the five characters fit in monohybrid segregation ratio. Chi square analysis revealed non-significant differences for all the morphological characters.

The F_2 showed 58, 117 and 43 plants with yellow, light yellow and cream coloured petals, respectively. They fit in 1:2:1 ratio, showing incomplete dominance. Similar studies conducted by Fyson (1908) revealed the same results wherein, he found F_1 was yellow and F_2 segregated sometimes into yellow, pale yellow and white and sometimes into full yellow and white only. Fletcher (1907) reported that yellow was dominant over white. Leake (1911a and 1911b) and Leake and Ramprasad (1914) reported that there are two allelomorphic pairs of factors involved in the crosses of full yellow with pale and white and it was confirmed by Hutchinson (1931).

Regarding petal spot colour, the F_2 plants showed 62, 105 and 51 plants with red, purplish red and purple petal spot colour, respectively. This also fits into 1:2:1 ratio. Soregaon

(2004) reported F_1 between *G. herbaceum* var. DDhc-11 and *G. anomalum* was intermediate with purplish red petal spot colour. The results observed here revealed purplish red petal spot colour as a codominant character. Similar studies were conducted by Harland and Atteck (1941a, 1941b) where results corroborate the present results. When anther colour was considered, it fitted in 3:1 ratio revealing it as a complete dominance and recessive relationship. Among F_2 plants, 152 plants showed yellow anthers similar to DDhc-11, whereas 66 plants showed cream anther similar to *G. anomalum* which corroborate with the results of Harland *et al.* (1941a).

In case of filament colour, the F_2 plants segregated in 3:1 ratio with 172 having cream yellow filament and 46 plants having purple filament, respectively. The results revealed that the filament colour shows dominance-recessive relationship. The purple filament, a characteristic of *G. anomalum* is governed by recessive gene which is in agreement with the results of Leake and Ramprasad (1914). In respect of bract type, plant numbers 68, 102 and 48 plants shown normal, intermediate and frego bracts, respectively. Studies conducted by Jones and Andries (1969) also revealed the recessive nature of frego bract character, which is in agreement with the present results.

5.2 GENETIC INTROGRESSION OF YIELD AND YIELD CONTRIBUTING CHARACTERS

Mere introgression of superior fibre qualities from wild species into cultivated species background may not fulfill the desiredness of introgression breeding, unless there is an increase in yield potential of such lines. The plant with high halo length also recorded high values for various yield and yield contributing characters like number of bolls per plant, seed cotton yield per boll weight, seed index, lint index, GOT etc. The data indicated the possibility of enhancing not only fibre properties but also yield and yield contributing characters from such interspecific crosses. A brief discussion in this regard is given in the following paragraphs.

5.2.1 Number of bolls per plant

The mean boll number of selected F_2 plants was significantly superior over the mean boll number of DDhc-11. Plant number 45 and 26 showed very high increase in the boll number when compared to all other plants. The standard deviation value of 48.64 shows the higher amount of variability among the F_2 plants. In heterosis studies for yield components Kajjidoni *et al.* (2004), Nirania *et al.* (2004), Sambamurthy *et al.* (2004), Siva Prasad *et al.* (2004) and Tangaraj *et al.* (2004) also observed significant increase in number of bolls per plant.

5.2.2 Seed cotton yield

Superiority of selected F_2 plants on the female parent DDhc-11 has been recorded but it was not significant. The standard deviation was 62.61 g, which is very high reflecting high variability. Out of all the selected F_2 plants, plant number 45 and 129 recorded maximum seed cotton yield indicating the possibility of improvement through interspecific hybridization. Similar increase in number of bolls per plant was also observed by Patel and Mehta (1990) and Kajjidoni (1982). In the study by Veeresh Gowda (2004) significant seed cotton yield increase of recombinant plants over check was observed in the cross between *G. herbaceum* var. Jayadhar and *G. barbadense* var. BCS-23. Superiority of seed cotton yield per plant in interspecific crosses was also reported by Tuteja and Singh (2001), Neelam Dheva *et al.* (2002) and Kharande *et al.* (2004).

5.2.3 Boll weight

The mean boll weight of selected F_2 plants was 1.35 g, which was inferior to the mean boll weight of DDhc-11 (2.01 g). The standard deviation of these F_2 plants was 0.37 g, which is not very high when compared to DDhc-11 (0.21 g). As the male parent *G. anomalum* is a lintless parent with a tapered boll shape, there is reduction in boll size in turn leading to reduction in boll weight. Patel *et al.* (2004) reported that there is reduction in per boll weight and cultivated varieties were used as female parents and multispecies derivatives were used as male parent.

5.2.4 Seed index

The mean seed index of F_2 plants was superior over DDhc-11, but it was not significant. The standard deviation of F_2 plants was 0.32, which is double the standard deviation of DDhc-11 indicating the presence of high variability in F_2. Veeresh Gowda (2004) observed significant increase in seed index over check in interspecific crosses. Bhatade and Rajeswari (1994) also reported superiority of recombinants over parents. Marani (1967 and 1968) and Amalraj (1989) reported superiority for seed index in interspecific hybrids. Anon. (2002) and Neelam Dheva *et al.* (2002) also reported similar results.

5.2.5 Ginning out turn

The ginning out turn of selected F_2 plants was significantly inferior to the DDhc-11. However, plant number 158 showed highest ginning out turn value (34.6%) among all the selected plants and eight F_2 plants showed higher ginning out turn values indicating the possibility for improvement in next generations. The standard deviation of these selected F_2 plants was very high (4.02%), which shows the maximum extent of variability existing for this character. These results are in confirmity with the results of Kharande *et al.* (2004), who reported reduction in GOT in the interspecific diploid cottons. Although, there is reduction in GOT, the increased GOT in some plants indicates that back crossing should be practiced along with selection to improve GOT.

5.2.6 Lint index

The lint index of F_2 is also inferior to DDhc-11. Reduction in lint index was also observed by Zhang Jinfa *et al.* (1994). However, plant number 74 and 138 recorded highest lint index. The standard deviation of F_2 plants is 0.61, which is very high indicating the presence of high variability for this character.

5.3 GENETIC INTROGRESSION OF FIBRE PROPERTIES

5.3.1 Halo length

The enormous variation (14.44 to 26.33 mm) was observed for halo length among F_2 plants and as many as 17 plants were having halo length of more than 22.00 mm. Similar studies done by Soregaon *et al.* (2004) and Veeresh Gowda (2004) also indicated high variation for halo length in recombinant plants in the cross between G. hirsutum var. DS-28 and *G. barbadense* var. SB(YF)-425. Manjula *et al.* (2004)observed wide variation 18.3 to 34.0 mm in the introgressed derivatives.

5.3.2 Estimation of fibre properties under HVI

The selected F_2 plants showed improved fibre properties over DDhc-11, thereby indicating the feasibility of ameliorating the fibre properties using wild relative *G. anomalum.*

5.3.2.1 Fibre length

The mean fibre length of selected F_2 plants was superior over DDhc-11 but it was not significant. The standard deviation value of 1.59 is 2 to 3 times more than that of DDhc-11 (0.39). Eleven plants were having fibre length of more than 24 mm, reflecting the improvement in the fibre length and scope to improve this character. Tuteja and Singh (2001), Modi *et al.* (1999), Naphade *et al.* (2004) and Kharande *et al.* (2004) reported superiority for fibre length in interspecific crosses.

5.3.2.2 Fibre strength

The mean fibre strength of selected F_2 plants (21.01 g/tex) was significantly superior over DDhc-11. The standard deviation value 1.81 g per tex of these plants indicates large amount of variability for this character. Most of the plants showed fibre strength values of higher than DDhc-11. Two plants with fibre strength of more than 24 g per tex were also isolated. Significant superiority for fibre strength was also reported by Zhang Jinfa *et al.* (1994), Chavan *et al.* (1999), Kharande *et al.* (2004) and Thangaraj *et al.* (2004).

5.3.2.3 Fibre elongation

The mean fibre elongation of F_2 plants was found significantly inferior to DDhc-11. The highest fibre elongation was recorded by plant number 26 (7.5) followed by plant number 136 (7.4) and 130 (7.4).

5.3.2.4 Fibre fineness

The mean micronaire of the F_2 plants was significantly lower to DDhc-11. In the present study, many plants with low micronaire value have been isolated. Plant number 203 showed lowest micronaire value (2.4 µg/inch) followed by plant number 17 (3.0 µg/inch). Results indicate that the fibre fineness of desi cottons has improved significantly. Tuteja and Singh (2001) and Naphade *et al.* (2004) observed similar results to fineness. The results corroborates the findings of Rao and Reddy (2001), Rajarathinam *et al.* (1993), Dheva and Potdukhe (2002), Ganeshan and Raveendran (2004)

5.3.2.5 Uniformity ratio

The mean uniformity ratio of the F_2 plants was superior to the mean uniformity ratio of DDhc-11. The highest uniformity ratio was recorded by two plants 34 and 17 (54.00%).

5.3.2.6 Fibre maturity ratio

The mean maturity ratio of F_2 plants was superior over mean maturity ratio of DDhc-11, but it was not significant.

5.4 MOLECULAR MARKERS ANALYSIS

Molecular markers are powerful tools to analyse genetic relationship and genetic diversity. A number of marker systems such as RAPD, AFLP, SSRs, ISSRs *etc*. are available to use in this endeavour. Restriction fragment length polymorphism (RFLPs) can be used, but they are expensive and time consuming. Among the several molecular techniques, random amplified polymorphic DNA (RAPD) markers (Williams *et al*., 1990) based on polymerase chain reaction (PCR) were shown to provide a high level of resolution equivalent to RFLPs for determining genetic relationship (Hallden *et al*., 1994). The technical simplicity and speed of RAPD methodology is a principal advantage (Gepts, 1993). Estimation of genetic similarity based on RAPDs have been obtained for rice (Cho *et al*., 1999), wheat (Liu *et al*., 1999), maize (Hahn *et al*., 1995), barley (Strelachenko *et al*., 1999) and several other crops.

RAPD markers have been successfully used to discriminate intra and interspecific genotypes in cotton (Iqbal *et al*., 1997; Multani and Lyon, 1995; Tatineni *et al*., 1996; Pawan Kumar *et al*., 2003; Soregaon *et al*., 2004; Veeresh Gowda *et al*., 2004; Rana and Bhat, 2004; Mehetre *et al*., 2004 and Hanchinal *et al*., 2004). In the present study RAPD was used to study genetic introgression among the F_2 plants derived from interspecific cross.

5.4.1 Genetic diversity

The dice similarity coefficient and cluster analysis revealed that there is similarity ranging from 60 to 82 per cent between the parents and the F_2 plants. The cluster diagram revealed that the male parent formed separate cluster with rest of the F_2 plants. It indicates that the nine plants were more similar to the DDhc-11 parent. The similarity coefficient (60.00%) between *G. anomalum* and the rest indicates that there is a partial homology between *G. anomalum* and DDhc-11. As DDhc-11 is having A1 genome and *G. anomalum* is having B1 genome, the *G. anomalum* is having partial homology to DDhc-11 parent. The prominent monomorphic fragments observed among the F_2 plants and parents indicates the common ancestral properties of these species. OPP-12 was most informative with seven polymorphic fragments (100%). OPC-12 was least informative with only 2 polymorphic fragment. The diversity is hypothesized to originate from interspecific cross derivatives, which is in agreement with the earlier reports (Vafai – Tabar *et al*., 2003). An average of 26.7 out of 35 specific bands came from female parent DDhc-11, whereas only 6.7 out of 18 bands came from male parent *G. anomalum* and 14 bands showed monomorphism. Plant number 16 recorded highest contribution (91.4%) of specific bands from female parent followed by 80 per cent by plant numbers, 158, 203 and 26. Plant number 17 showed highest contribution (68.7%) from male parent.

5.4.2 Identification of marker linked to fibre strength

A 300 bp fragment amplified from OPZ-19 had been noticed only in groups having fibre strength 23.0, 24.0 and 25.0 g per tex plants along with *G. anomalum* and F_1. The repeatability of this marker had been checked by taking each group in three replications. Again on repetition also this marker amplified in high fibre strength lines. So, this marker may be used in suitable population for linkage study.

5.5 FUTURE LINE OF WORK

1. Development of near isogenic lines for fibre strength
2. Validation of the putative marker identified in the present study and some more extensive research for molecular markers linked to fibre strength.
3. Further handling of the selected segregants to identify high yielding desi type cotton with improved fibre properties.
4. Continuation of introgression hybridization to realize the objective as mentioned in (3) above.

VI. SUMMARY

Cotton is one of the most important fibre crop in the world, which is having special significance in terms of commercial importance, it is an inevitable source of natural fibre in the textile industry throughout the world. It is a prime raw material (85%) of textile industry, which provides employment to millions of people in the world under various activities.

In the present study, *G. herbaceum* var. DDhc-11 was used as female parent and *G. anomalum* was used as male parent. *G. herbaceum* var. DDhc-11, which is cultivated diploid variety is having some constraints with respect to fibre properties. In order to improve fibre properties of DDhc-11, *G. anomalum* a diploid wild relative was used as male parent the F_2 population was used in the present study. The investigation was conducted at Agricultural Research Station, Dharwad Farm during 2002-03 with the aim to develop the recombinant plants with superior fibre properties.

Genetic introgression studies for morphological characters

Distinguishable morphological qualitative characters such as petal colour, petal spot colour, anther colour, filament colour and bract type were studied. All these characters fitted in to monohybrid ratios.

Genetic introgression studies of yield and yield contributing characters

Plants, which possessed higher halo length and sufficient lint were subjected for detailed study. Observations on characters like number of bolls per plant, seed cotton yield per plant, boll weight, seed index, ginning out turn and lint index were recorded. The mean number of bolls per plant and GOT of selected F_2 plants showed significant superiority and inferiority, respectively over the female parent DDhc-11. Whereas, seed cotton yield and seed index showed non-significant superiority and boll weight and lint index showed non-significant inferiority.

Genetic introgression of fibre properties

Twenty two plants were analysed for fibre properties for characters like fibre strength, fibre fineness and elongation percentage F_2 plants showed had shown significant superiority over DDhc-11. For fibre length, uniformity ratio and maturity ratio numerical superiority was observed.

Genetic diversity through molecular markers

Genetic diversity analysis

Eleven primers that had shown polymorphism were used for genetic diversity study. Out of 65 fragments, 43 were found to be polymorphic, which indicated the presence of genetic diversity among F_2 plants. Out of 5.4 fragments per primer amplified, 3.58 fragments per primer were polymorphic. The cluster diagram and dice similarity coefficients indicated 60 to 82 per cent similarity between selected F_2 plants and the parents. The *G. anomalum* formed separate cluster. The molecular markers indicated an average of 76.4 per cent of specific bands from DDhc-11 and only 42.3 per cent of specific bands from *G. anomalum*.

Identification of markers linked to fibre strength

Based on fibre strength values, 22 plants were categorized into nine groups representing the fibre strength from 17 to 25 g per tex. These F_2 plants, along with parents and F_1 were screened using RAPD primers, OPZ-19 amplified a 300 bp amplicon, which was present only in groups representing 23, 24 and 25 g per tex along with P_2 and F_1, but was absent in other groups indicating its association with fibre strength.

VII. REFERENCES

ABDEL GHANY, A., ABDEL GHANY AND ZAKI, E. A., 2003, DNA sequences of RAPD fragments in the Egyptian cotton *Gossypium barbadense*. *African Journal of Biotechnology*, **2** : 129-132.

AMALRAJ, S. F. A., 1989, Combining ability studies on *Gossypium hirsutum* × *Gossypium barbadense* hybrids. *Indian Journal of Agricultural Research,* **23**(2) : 65-69.

AMIN, K. C., 1940, Interspecific hybridization between Asiatic and new world cottons. *Indian Journal of Agricultural Sciences*, **10** : 404-413.

ANONYMOUS, 2002, Level of heterosis for quantitative traits in upland cotton. *Journal of Indian Society for Cotton Improvement,* **27**(2) : 200-203.

ANONYMOUS, 2005a, *Annual Report of Project Co-ordinator*, All India Co-ordinated Cotton Improvement Project, pp. 1-5.

ANONYMOUS, 2005b, Training Manual on DUS test in Cotton with Reference to PPV and FR Legislation, 2001, All India Co-ordinated Cotton Improvement Project, CICR, Coimbatore, Tamil Nadu, pp. 134-135.

APPATHURAI, R., PONNAIYA, B. W. X., SANTHANAM, V. AND RAMAN, V. S., 1964, Interspecific hybridisation in *Gossypium* – study of *G. arboreum* x *G. anomalum* back cross hybrids. *Madras Agricultural Journal*, **51** : 358.

ARUTYUNOVA, L. G. AND VOLKOVA, L. A., 1987, Cytological characterization of triploid and hexaploid cotton hybrids. *Semenovodstva Kolapchanika*, **20** : 28-43.

ASHOK DONGRE, VILAS PARKHI AND SANTHOSH GAHUKAR, 2004, Characterization of cotton (*G. hirsutum*) germplasm by ISSR, RAPD markers and agronomic values. *Indian Journal of Biotechnology*, **3** : 388-393.

BALLS, W. L., 1906, Studies in Egyptian cotton in 'yearbook khediv, agvic SOC for 1906', pp : 29-89.

BHATADE, S. S. AND RAJESWARI, S. R., 1994, Heterobeltiosis and standard heterosis for yield and quality characters in some *Gossypium hirsutum* L. crosses. *Madras Agricultural Journal*, **81** : 34-35.

BRAR, K. S., SANDHU, B. S., GOSSAL, S. S. AND GIRHOTRA, R. P., 1999, *In vitro* interspecific hybridization in *Gossypium* species. *Journal of the Indian Society for Cotton Improvement*, **24**(1) : 23-26.

CARNELL, R. G. AND DAVIS, D. D., 1993, Characterization of *G. hirsutum* x *G. barbadense* breeding lines using molecular markers. In *Proceedings of Belt wide Production Conference,* Ed. D. J. Herber, National Cotton Council of America, pp. 1551-1553.

CHAVAN, M. K., SHEKAR, V. B., GOLHAR, S. R., GITE, B. D. AND RAJPUT, N. R., 1999, Heterosis studies in interspecific crosses of *G. arboreum* and *G. herbaceum*. *Journal of Soils and Crops*, **9**(2) : 195-198.

CHO, Y. C., SHIN, Y. S., AHU, S., GREGORIA, G. B., KANG, K., H. O., BRAR, D. AND MOON, H., 1999, DNA fingerprinting of rice cultivars using AFLP and RAPD markers. *Korean Journal of Crop Science*, **44** : 26-31.

CULP, T. W. AND HARRELL, D. C., 1980, Registration of extra-long staple cotton germplasm (REG. No. GP 150 to GP 154). *Crop Science*, **20**(2) : 291.

DARK, S. O. S., 1960, Plant hairiness and staple cotton, *Empirical Cotton Growing Review*, **37** : 266-269.

DAVIS, D. D., 1993, Registration of six upland type parental R-lines for hybrid cotton. *Crop Science*, **33**(6) : 1428-1429.

DESHPANDE, L. A., KOKATE, R. M., KULKARNI, U. G. AND NEVKAR, Y. S., 1992, Cytomorphological studies in induced tetraploid *G. arboreum* (4n=52) and new interspecific hybrid between 4n *G. arboreums* x *G. hirsutum* L. In *Proceedings of the First Vasant Rao Naik Memorial National Seminar on Agricultural Sciences Cotton Development,* held between 5th and 6th December at Nagpur India, pp. 38-47.

DHEVA, N. G. AND POTDUKHE, N. R., 2002, Studies on variability and correlations in upland cotton for yield and its components. *Journal of Indian Society for Cotton Improvement*, pp. 148-152.

DOUGLAS, C. R. AND BROWN, M. S., 1971, A study of the triploid and 3x-I aneuploid plants in the genus *Gossypium*. *American Journal of Botany*, **58** : 65-71.

EL-HATTAB, H. E., RADWAN, S. R. H. AND EMARA, E. H., 1974, Potentialities of gene exchange in selection programs for earliness in some crosses between Egyptian and upland cottons. *Agricultural Research Review*, **52**(8) : 65-74.

FENG, C. F., 1935, A genetic and cytological study of hybrids of Asiatic and American cotton. *Journal of Agricultural Research. National Central University*, Nanking, **1** : 77-87.

FLETCHER, F., 1907, Mendelian heredity in cotton. *Journal of Agricultural Sciences*, **2** : 281-282.

FYSON, P. F., 1908, Some experiments in the hybridizing of Indian cottons. *Memorandum of Department of Agriculture and Indian Botany*, **2**(6) : 1-29.

GALANOPOULOU-SENDOUCA, S. AND ROUPAKIAS, D., 1999, Performance of cotton F_1 hybrids and its relation to the mean yield of advanced bulk generations. *European Journal of Agronomy*, **11**(1) : 53-62.

GANESHAN, D., 1946, Interspecific hybridization in cotton. *Third Conference on Cotton Growers Problems*, India, I. C. C. C., pp. 80-86.

GANESHAN, K. N. AND RAVEENDRAN, T. S., 2004, Genetic variability studies on fibre quality traits and seed cotton yield in F_2 and F_3 generations of cotton (*G. hirsutum* L.). *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 190-192.

GEPTS, P., 1993, The use of molecular and biochemical markers in crop evolution studies. In *Evolutionary Biology*, Ed. M. K. Hecht, Plenum Press, New York, **27** : 51-94.

GILL, M. S. AND BAJAJ, Y. P. S., 1984, *In vitro* production of interspecific hybrid embryos in cotton. *Current Science*, **53**(4) : 102-104.

GOMES, M., KULOTHUNGAN, G., MEHETRE, S. S. AND EAPAN, S., 2004, Analysis of genetic relatedness in *Gossypium* species using RAPD. *Indian Journal of Biotechnology*, **3** : 41-46.

GURURAJAN, K. N. AND SUNDAR, S., 2004, Yield component analysis in American Cotton (*Gossypium hirsutum* L.). *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 201-204.

HAHN, V., BALNKENHOM, K., SCHWALL, M., MELCHINGER, A. E., MALECOT, G. AND JACCARD, P., 1995, Relationships among early European maize inbreds II. Genetic Diversity revealed with RAPD markers and comparison with RFLP and pedigree data. *Maydica*, **40** : 299-310.

HALLDEN, C., NILSSON, N. O., RADING, I. M. AND SALE, T., 1994, Evaluation of RFLP and RAPD markers in comparison of *Brassica napus* breeding lines. *Theoretical and Applied Genetics*, **88** : 123-128.

HANCHINAL, R. R., NADAF, H. L., NISHANI, S. S., MOTAGI, B. N., MADHUSUDAN, K. AND VIJAYKUMAR, A. G., 2004, Varietal identification and hybrid purity testing through RAPD markers in cotton (*Gossypium* sp.). *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 406-410.

HARLAND, S. C. AND ATTECK, O. M., 1941a, The genetics of cotton XVIII. Transference of genes from diploid North American wild cottons to tetraploid new world cottons. *Journal of Genetics*, **42** : 1-19.

HARLAND, S. C. AND ATTECK, O. M., 1941b, The genetics of cotton XIX. Normal alleles of the crinkled mutant of *G. barbadense* L. differing in dominance potency and an experimental verification of Fisher's theory of dominance. *Journal of Genetics*, **42** : 21-47.

HUTCHINSON, J. B., 1931, The genetics of cotton Part IV. The inheritance of corolla colour and petal size in Asiatic cottons. *Journal of Genetics*, **24** : 325-353.

HUTCHINSON, J. B., 1951, Intraspecific differentiation in *G. hirsutum*. *Heredity*, **5** : 161-193.

HUTCHINSON, J. B., 1959, The application of genetics of cotton improvement, Cambridge University Press, London.

HUTCHINSON, J. B., SILOW, R. A. AND STEPHENS, S. G., 1945, Progress report from experimental stations Trinidad. *Empirical Cotton Growing Review*, 1941-1942.

HYER, A. H., 1973, Comparative performance of isogenic glanded and glandless cotton lines. *Agronomy Abstracts*, p. 7.

IQBAL, M. J., AZIZ, N., SAEED, N. A., ZAFAR, Y. AND MALIK, K. A., 1997, Genetic diversity evaluation of some elite cotton varieties by RAPD analysis. *Theoretical and Applied Genetics*, **94**(1) : 139-144.

IYANAR, K., RAVIKESAVAN, R., SUBRAMANIAN, A., RAVEENDRAN, T. S. AND VINDHIYA VARMAN, P., 2004, Studies on identificaiton of superior introgressed lines for yield, quality and biotic stress tolerance in upland cotton (*G. hirsutum* L.). *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 292-295.

JONES, J. E. AND ANDRIES, J. A., 1969, Effect of frego bract on the incidence of cotton boll rot. *Crop Science,* **9** : 426-428.

KACHAPUR, R. M., 2001, *In vivo* and *in vitro* interspecific cross recovery studies in cotton (*Gossypium* spp.). *M. Sc. (Agri.) Thesis,* University of Agricultural Sciences, Dharwad.

KADAPA, S. N. AND GANGA PRASAD, N., 1965, Utilization of perennial tetraploid cottons in improving the annual *G. hirsutum* variety. *Indian Cotton Journal*, **14**(5) : 286-290.

KAJJIDONI, S. T., KHADI, B. M. AND SALIMATH, P. M., 2004, Relative contribution of component traits of seed cotton yield in intra arboreum GMS based and their conventional hybrids in desi cotton. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 220-222.

KATAGERI, I. S. AND KADAPA, S. N., 1989, Heterosis and gene action for yield and contributing characters in interspecific hybrids (*G. hirsutum* x *G.*

barbadense). *The Indian Journal of Genetics and Plant Breeding*, **49**(1) : 107-111.

KATAGERI, I. S., KHADI, B. M. AND VAMADEVAIAH, H. M., 1999, *In vitro* inovuloembryo culture in cotton (*Gossypium* spp.). *Journal of Indian Society for Cotton Improvement*, **24**(2) : 95-107.

KATAGERI, I. S., KHADI, B. M., IMMADI, S., SUVARNA, B., MANJULA, S. M. AND BHANDARI, A., 2004, In vitro protocol for induction of colchiploidy in diploid cotton. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 427-432.

KATAGERI, I. S., KHADI, B. M., MANOHAR, K., SOREGAON, C. D., VAMADEVAIAH, H. M., MANJULA, S. AND BADIGANNAVAR, A. M., 2003, Recombinant lines in cotton (*Gossypium* spp.). *Paper Presented at World-Cotton Research Conference III* held at Capetown, South Africa, 9-13 March.

KATARKI, B. H., 1971, Varalaxmi, a high yielding hybrid cotton of quality. *Indian Farming*, **21**(8) : 35-36.

KATARKI, B. H., 1981, *Annual Report*, DCH-32, high yielding interspecific hybrid, University of Agricultural Sciences, Bangalore.

KHADI, B. M., 1996, A new interspecific cotton hybrid for Southern Zone. *National Seminar on Centenary Celebration of Cotton in India*, Souvenir, p. 10.

KHAN, S. A., HUSSAIN, ASKARI, STEWART, J. M., MALIK, K. A. AND ZAFAR, Y., 2000, Molecular phylogeny of *Gossypium* species by DNA fingerprinting. *Theoretical and Applied Genetics,* **101**(5-6) : 931.

KHARANDE, S. S., WANDHARE, M. R., LADOLE, M. Y., WAKODE, M. M. AND MESHRAM, L. D., 2004, Heterosis and combining ability studies in interspecific diploid cotton hybrids for fibre quality parameters. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 270-273.

KIMBER, G. AND RILEY, R., 1963, Haploid angiosperms. *Botanical Review*, **29** : 480-537.

KNIGHT, R. L., 1956, The genetical approach to disease resistance in plants. *Empirical Cotton Growing Review*, **33** : 191-196.

KULKARNI, V. N. AND KHADI, B. M., 1998, Long linted *G. arboreum* for meeting textile industrial needs. *New Frontiers in Cotton Research : Proceedings of World Cotton Research Conference-II*, Athens, Greece, September 6-12.

KUMAR, P. R., RAVEENDRAN, T. S. AND KRISHNAN, V., 2000, Genetic diversity and association studies for fibre characters in cotton (*G. hirsutum* L.). *Journal of Indian Society for Cotton Improvement*, **14**(1) : 111-118.

LEAKE, H. M., 1911a, Studies on Indian cotton. *Journal of Genetics*, **1** : 205-272.

LEAKE, H. M., 1911b, Experimental studies in Indian cotton. *Proceedings of Royal Society Ser. B.*, **83** : 447-451.

LEAKE, H. M. AND RAMPRASAD, 1914, Studies on Indian cottons I. The vegetative characters. *Memorandum of Department of Agriculture and Indian Botany*, **6** : 115-150.

LEE, J. A., 1970, The origin of haploid/diploid twinning in cotton. *Crop Science*, **10** : 453-463.

LIU, Z. Q., DEI, Y. AND PU, Z. J., 1999, Relationship between hybrid performance and genetic diversity based on RAPD markers in wheat. *Plant Breeding*, **118** : 119-123.

MANICKAM, A., PARAMESWARI, C., INDUMATHI, M. AND CHANDRAN, A. H., 2002, DNA fingerprinting in cotton (*Gossypium* spp. L.) based on RAPD markers for varietal differentiation. *Research on Crops*, **3**(3) : 635-642.

MANJULA, S. M., KHADI, B. M., PAWAR, S. V., IMMADI, S. AND KATAGERI, I. S., 2004, Improvement of *G. herbaceum* through introgression breeding. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 300-302.

MANOHAR, K. K., 2003, Effect of blooming dates on yield, yield contributing and fibre quality parameters and introgression of fibre qualities in *Gossypium* spp. *M. Sc. (Agri.) Thesis*, University of Agricultural Sciences, Dharwad.

MARANI, A., 1967, Heterosis and combining ability in intraspecific and interspecific crosses of cotton. *Crop Science*, **7** : 519-520.

MARANI, A., 1968, Heterosis and combining ability of quantitative characters in interspecific crosses of cotton. *Crop Science*, **8** : 299-303.

McCARTY, J. C. Jr., JENKINS, J. N. AND TANG, B., 1995, Primitive cotton germplasm : variability for yield and fibre traits. *Technical Bulletin*, Mississippi Agricultural and Forestry Experiment Station, No. 202, p. 8.

MEHETRE, S. S., SANTOSH GAHUKAR AND NAIK, R. M., 2004, RAPD marker inheritance in segregating generation of interspecific cross. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 403-405.

MEHTA, N. P., BADAYA, S. N. AND PATEL, G. S., 1988, A modified hybrid seed production technique for Asiatic cotton. *Cotton Development*, **13**(1) : 21-22.

MEHTA, N. P., BASU, A. K. AND MANDOLI, K. C., 1985, Rainfed cotton production technology-crop improvement. In : *Proceedings of National Seminar on Rainfed Cotton Production Technology*, May 24, Maharashtra Agricultural University, Parbhani, pp. 9-24.

MEREDITH, W. R., 1993, Registration of 'MD5lne' cotton. *Crop Science*, **33**(6) : 1415.

MEYER, V. A. AND MEREDITH, W. R., 1978, New germplasm from crossing Upland Cotton (*G. hirsutum*) with *G. tomentosum*. *Journal of Heredity*, **69** : 183-187.

MEYER, V. G., 1974, Interspecific cotton breeding. *Economic Botany*, **28** : 56-60.

MODI, N. D., PATEL, J. C., PATEL, D. H., MAISURIA, A. T. AND PATEL, U. G., 1999, Heterosis and combining ability for yield and fibre quality in desi cottons. *Journal of Indian Society for Cotton Improvement*, **24**(2) : 132-134.

MULTANI, D. S. AND LYON, B. R., 1995, Genetic fingerprinting of Australian cotton cultivars with RAPD markers. *Genome*, **38** : 1005-1008.

MURTHY, J. S. V. S., REDDY, D. M. AND REDDY, K. H. G., 1995a, Studies on the nature of genetic divergence in upland cotton (*G. hirsutum* L.). *Annals of Agricultural Research*, **16**(3) : 307-310.

MURTHY, J. S. V. S., REDDY, D. M. AND REDDY, K. H. G., 1995b, Genetic divergence for lint characters in upland cotton (*G. hirsutum* L.). *Annals of Agricultural Research*, **16**(3) : 357-359.

NAPHADE, P. S., KHARDES, R. P., POTDUKHE, N. R. AND MESHRAM, L. D., 2004, Heterosis and combining ability for quality attributes in inter-varietal crosses of diploid cotton. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 267-269.

NARAYANAN, S. S., SINGH, J. AND VARMA, P. K., 1984, Introgressive gene transfer in *Gossypium*, goals, problems strategies and achievements. *Cotton Fibre Tropics*, **39**(4) : 123-135.

NEELAM DHEVA, POTDUKHE, N. R. AND PATIL, V. T., 2002, Heterosis for seed cotton yield and other morphological characters in *G. hirsutum* L. *Journal of Cotton Research and Development*, **16**(2) : 165-167.

NIE, Y. C., ZUO, K. J., ZHANG, X. L., FENG, C. D. AND LIU, J. L., 2000, Application of RAPD markers in differentiating germplasm lines from 4x (*Gossypium arboreum* × *G. anomalum*) × *G. hirsutum*. *Scientia Agricultura Sinica*, **33**(5) : 25-29.

NIRANIA, K. S., CHHABRA, B. S., JAIN, P. P., YADAV, J. P. S. AND YAGYA DUTT, 2004, Identification of superior combining parents for yield and quality traits for development of GMS based hybrids in upland cotton. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 223-225.

NIU, Y. Z., ZHANG, Y. G., GUO, B. D. AND HUA, S. L., 1998, Research on creating new germplasm lines through interspecific hybridization in *Gossypium*. *China Cottons*, **25**(1) : 16-17.

OMEL-CHENKO, V. M., ABDULLAEV, A. A. AND LIKHODZIEVSKAYA, A. A., 1979, Economically useful recombinants after crossing wild and cultivated cotton species. *Referativnyi Zhurnal*, **1** : 65-73.

PANDYA, P. S. AND PATEL, C. T., 1956, Review of further progress in inter-specific hybridization work at Surat. *Proceedings of 7th Conference on Cotton Growing Problems,* India, pp. 9-14.

PATEL, J. N. AND DESAI, K. B., 1963, Improvement of Surti cotton through hybridization with Iranian *herbaceum*. *Indian Cotton Growing Review*, **17**(3) : 186.

PATEL, K. G., PATEL, U. G., MAISURIA, A. T. AND CHHIMPI, B. G., 2004, Heterosis and combining ability in crosses involving multispecies derivatives. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 274-277.

PATEL, U. G. AND MEHTA, N. P., 1989, Combining ability studies on yield and yield components on *desi* cotton (*G. herbaceum*). *Journal of Indian Society for Cotton Improvement*, **14** : 45-51.

PATEL, U. G. AND MEHTA, N. P., 1990, Heterosis in multiple environments for seed cotton yield and major yield components in *G. herbaceum*. *Journal of Indian Society for Cotton Improvement*, **15** : 1-5.

PAWAN KUMAR, SINGH, K., VIKAL, Y., RANDHAWA, L. S. AND CHAHAL, G. S., 2003, Genetic diversity studies of elite cotton germplasm lines using RAPD markers and morphological characteristics. *The Indian Journal of Genetics and Plant Breeding*, **63**(1) : 5-10.

PERCIVAL, E. AND KOHEL, R. J., 1990, Distribution collection and evaluation of *Gossypium*. *Advances in Agronomy*, **44** : 225-228.

PHILLIPS, L. L., 1974, Cotton (*Gossypium*). In *Handbook of Genetics,* Plenum Press, N. Y. and London, **12** : 111-133.

PURI, S. N. AND MEHETRE, S. S., 2001, Sustainable cotton production to meet future requirements of the industry. *National Seminar on Sustainable Cotton*

Production to Meet the Requirement of Industry, held on 3-4 October 2001 at Mumbai.

RAJARATHINAM, S. AND NADARAJAN, N., 1993, Multi-variate analysis of genetic divergence in upland cotton (*G. hirsutum* L.). *Journal of Indian Society for Cotton Improvement*, **18**(1) : 107-112.

RAJARATHINAM, S., NADARAJAN, N. AND SUKANYA SUBRAMANIAM, 1993, Genetic variability and association analysis in cotton. *Journal of Indian Society for Cotton Improvement*, **18** : 54-59.

RAJASHEKHAR, M. K., KATAGERI, I. S., KHADI, B. M. AND VAMADEVAIAH, H. M., 2003, *In vitro* culture of interspecific hybrid ovules and embryos of cotton (*Gossypium* spp.). *Plant Cell Biotechnology and Molecular Biology*, **4**(3&4) : 151-156.

RAMACHANDRAN, C. K., KRISHNAMURTHY, J. AND PETER, S. D., 1964, Recent advances in interspecific hybridization work involving wild species of cotton in Madras. *Indian Cotton Growing Review*, **18**(4) : 248-257.

RANA, M. K. AND BHAT, K. V., 2002, Genetic diversity analysis in Indian diploid cotton (*Gossypium* spp.) using RAPD markers. *The Indian Journal of Genetics and Plant Breeding*, **62**(1) : 11-14.

RANA, M. K. AND BHAT, K. V., 2004, Role of molecular markers in fingerprinting of varieties and parents of hybrids in the context of IPR issues. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 370-372.

RAO, G. N. AND REDDY, M. S. S., 2001, Studies on heritability and variability for yield and its components in cotton (*Gossypium hirsutum* L.). *Journal of Cotton Research and Development*, **15**(1) : 84-86.

RICHMOND, T. R., 1951, Procedure and methods of cotton breeding with special reference to American cultivated species. *Advances in Genetics*, **4** : 213-245.

ROHLF, F. J., 1998, NTSYS-PC numerical taxonomy and multivariate analysis, version 2. 0. *Applied Biostatics* Inc., New York.

SAGHAI-MAROOF, M. A., SOLIMAN, K. M., JORGENSEN, R. A. AND ALLARD, R. W., 1984, Ribosomal DNA spacer length polymorphism in barley: Mendelian inheritance, chromosomal location and population dynamics. *Proceedings of National Academy of Sciences*, (USA) **81** : 8014-8018.

SAMBAMURTHY, J. S. V., RATNAKUMARI, S., CHAMUNDESHWARI, N. AND SHIVA REDDY, K. V., 2004, Studies on nature of genetic divergence in diploid cotton (*G. herbaceum* L.). *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 187-189.

SANTHANAM, V. AND KRISHNAMURTHY, R., 1961, Sujata - A new high spinning cotton variety. *Indian Farming*, **20**(2) : 5-7.

SHERZHANOVA, K., 1987, Ecologically distant hybridization. *Khlopkovodstova*, **2** : 27-29.

SINGH, P., 1998, "*Cotton Breeding*", Kalayani Publishers, New Delhi.

SINGH, P. AND NARAYANAN, S. S., 1991, Genetical improvement of *arboreum* cotton in India. *Journal of the Indian Society for Cotton Improvement*, **16** : 81-96.

SIVA PRASAD, U., CHENGA REDDY, V. AND NARISI REDDY, A., 2004, Combining ability studies for yield and yield components in American cotton. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 264-266.

SKOVSTED, A., 1935, Some new interspecific hybrids in genus *Gossypium*. *Journal of Genetics*, **30** : 447-463.

SOREGAON, C. D., 2004, Studies on genetic introgression in interspecific crosses of cotton. *M. Sc. (Agri.) Thesis*, University of Agricultural Sciences, Dharwad.

SOREGAON, C. D., KATAGERI, I. S., KHADI, B. M., VAMADEVAIAH, H. M. AND ANITHA, B., 2004, Genetic diversity analysis in recombinants derived from interspecific crosses of cotton (*Gossypium spp*) using RAPD markers. *Paper Presented at International Symposium On Strategies For Sustainable Cotton Production – A Global Vision* held at Dharwad, India, 23-25 November, 2004.

SREENIVASAN, S., 2001, Quality profile of popular hybrids and varieties and measures for augmentation. *National Seminar on Sustainable Cotton Production to Meet the Requirement of Industry*, held on 3-4 October 2001 at Mumbai.

SREERANGASWAMY, S. R. AND RAMAN, V. S., 1963, Studies on the morphology and cytology of *Gossypium herbaceum* x *G. anomalum* hybrids. *Indian Cotton Growing Review*, **17** : 353-359.

STALKER, H. T., 1980, Utilization of wild species for crop improvement. *Advances in Agronomy*, **33** : 11-17.

STEBBINS, G. L., 1958, The inviability weakness and sterility of interspecific hybrids. *Advances in Genetics*, **9** : 147-155.

STEPHENS, S. G., 1949, The cytogenetics of speciation in *Gossypium*. I. Selective elimination of the donor parent genotype in interspecific backcrosses. *Genetics*, **34** : 627-637.

STRELACHENKO, P., KOVALYOVA, O. AND OKUNO, K., 1999, Genetic differentiation and geographical distribution of barley germplasm based on RAPD markers. *Genetic Resources and Crop Evolution*, **46** : 193-205.

TATINENI, V., CANTRELL, R. G. AND DAVIS, D. D., 1996, Genetic diversity in elite cotton germplasm determined by morphological characteristics and RAPDs. *Crop Science*, **36**(1) : 186-192.

THANGARAJ, K., RAVEENDRAN, T. S. AND RAVIKESAVAN, R., 2004, Genetic studies for yield and fibre quality characters in the racial and wild derivatives of Gossypium with American bollworm resistance. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 205-208.

THOMBRE, M. V. AND MEHETRE, S. S., 1979, Note on haploids of the interspecific cross of a cotton. *Indian Journal of Agricultural Research*, **13** : 195-196.

THOMBRE, M. V. AND MEHETRE, S. S., 1981, Interspecific hybridization in genus *Gossypium* L. Cytomorphological studies in hybrid *G. hirsutum* haploid (2n=2x=26, Ah, Dh) and *G. thurberi* (2n=2x=26, D_1D_1). *Cytologia*, **46** : 291-299.

TUTEJA, O. P. AND SINGH, D. P., 2001, Heterosis for yield and its components in Asiatic cotton hybrids based on GMS system under varied environment. *Indian Journal of Genetics*, **61**(3) : 291-292.

VAFAIE-TABAR, M., CHANDRASHEKARAN, S., SINGH, R. P. AND RANA, M. K., 2003, Evaluation of genetic diversity in Indian tetraploid and diploid cotton (*Gossypium* spp.) by morphological characteristics and RAPDs. *The Indian Journal of Genetics and Plant Breeding*, **63**(3) : 230-234.

VAMADEVAIAH, H. M., KATAGERI, I. S., BHANDARI, A. AND KHADI, B. M., 2004, Identification of RAPD markers in genetic purity tests of DHH-11 intra hirsutum hybrid. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 389-391.

VEERESH GOWDA, R. P., 2004, Studies on genetic recombination in interspecific crosses of *Gossypium* spp. *M. Sc. (Agri.) Thesis*, University of Agricultural Sciences, Dharwad.

VEERESH GOWDA, R. P., KATAGERI, I. S., KHADI, B. M., VAMADEVAIAH, H. M. AND BHANDARI, A., 2004, Extra long staple *G. hirsutum* L. cottons derived from *G. hirsutum* L. × *G. barbadense* L. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 306-309.

VINITA, G. AND PHUNDAN SINGH, 2004, Cotton improvement through use of wild species in India. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 288-291.

WILLIAMS, J. G. K., KUBELIK, A. R., LIVAK, K. J., RAFALSKI, J. A. AND TINGEY, S. V., 1990, DNA polymorphism amplified by arbitrary primers are useful as genetic markers. *Nucleic Acids Research*, **18** : 6531-6535.

XIANLONG ZHANG, ZHONGXU LIN, DAOHUA HE , YICHUN NIE AND XIAOPING GUO, 2004, A genetic linkage map of tetraploid cotton constructed using SSRs, RAPDs and SRAPs. *Plant and Animal Genomes XII Conference.*

YADAV, M., RANADE, R., VAIDYA, V. J. AND GOPALA KRISHNA, T., 2001, Genetic purity determination of cotton hybrid DCH-32 by random amplified polymorphic DNA (RAPD). *Plant Varieties and Seeds*, **14**(1) : 35-40.

YINGZHI LU, PERCY, R. G., CANTRELL, R. G. AND JINFA ZHANG, 2004, A comprehensive linkage map using recombinant inbred line populations in cultivated tetraploid cottons. *Plant and Animal Genomes XII Conference.*

ZAHID MAHMOOD, ABDUL RAZAQUE SOOMRO, ASIA PERVEEN, MOHAMMAD QAUSER ALAM AND ALTAF AHMAD DASTI, 2004, Ancestors of *Gossypium hirsutum* : A molecular study. *International Symposium on Strategies for Sustainable Cotton Production – A Global Vision*, 23-25 November, 2004, held at University of Agricultural Sciences, Dharwad, pp. 392-397.

ZHANG JINFA, DENG ZHONG, SUN JIZHONG AND LIU JINLUN, 1994, Heterosis and combining ability in interspecific crosses between *G. hirsutum* and *G. barbadense*. *Journal of Huazhong Agricultural University,* **13**(1) : 9-14.

ZHANG, J., GUO, W. AND ZHANG, T., 2002, Molecular linkage map of allotetraploid cotton (*Gossypium hirsutum* L. X *Gossypium barbadense* L.) with a haploid population. *Theoretical and Applied Genetics*, **105** : 1166-1174.

ZHANG, T. Z., YUAN, Y. L., YU, J., GUO, W. Z. AND KOHEL, R. J., 2003, Molecular tagging of a major QTL for fibre strength in upland cotton and its marker-assisted selection. *Theoretical and Applied Genetics*, **106**(2) : 262-268.

Appendix I: Weather data at ARS, Dharwad Farm for the year 2004-05 and normal of 68 years

Months	Mean of 68 years (1936 to 2004)		2004-05	
	Rainfall (mm)	Rainy days	Rainfall (mm)	Rainy days
April	47.02	3	88.6	6
May	82.21	8	69.6	6
June	104.69	13	117.4	13
July	135.85	23	25.0	4
August	93.71	20	174.8	13
September	109.88	12	235.6	11
October	121.64	11	23.8	3
November	33.50	3	0.0	0
December	7.00	3	0.0	0
January	2.00	0	13.2	1
February	0.00	0	0.0	0
March	0.00	0	0.0	0
Total	**737.50**	**94**	**748.0**	**57**

Appendix II: Morphological characters of F_2 population

Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type	Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type
1.	Y	PR	C	CY	I	26.	LY	PR	Y	CY	N
2.	Y	PR	Y	CY	I	27.	C	P	Y	CY	I
3.	LY	R	Y	CY	N	28.	Y	R	Y	P	F
4.	C	PR	Y	CY	F	29.	LY	PR	C	CY	N
5.	Y	R	Y	P	I	30.	C	P	Y	CY	I
6.	LY	PR	Y	CY	I	31.	LY	P	Y	CY	F
7.	C	R	Y	CY	N	32.	LY	R	Y	CY	N
8.	Y	P	C	CY	I	33.	Y	PR	Y	CY	I
9.	LY	PR	Y	CY	F	34.	LY	PR	Y	CY	N
10.	C	P	C	CY	I	35.	C	R	Y	CY	F
11.	Y	R	Y	P	I	36.	LY	P	C	P	N
12.	LY	P	Y	CY	N	37.	LY	PR	C	CY	I
13.	C	PR	C	CY	I	38.	Y	PR	Y	CY	F
14.	LY	P	C	P	F	39.	LY	P	C	CY	F
15.	C	R	Y	CY	F	40.	LY	P	Y	P	N
16.	LY	P	Y	CY	N	41.	C	R	Y	CY	I
17.	LY	R	Y	P	N	42.	LY	R	Y	P	N
18.	Y	PR	Y	CY	I	43.	C	P	C	CY	F
19.	LY	P	Y	CY	I	44.	LY	PR	Y	P	I
20.	C	PR	Y	CY	N	45.	LY	P	Y	P	N
21.	Y	PR	Y	P	I	46.	C	P	Y	CY	I
22.	LY	P	C	CY	I	47.	LY	R	Y	P	I
23.	LY	P	Y	CY	N	48.	LY	P	C	CY	I
24.	C	PR	Y	CY	I	49.	C	PR	Y	CY	I
25.	Y	PR	Y	CY	I	50.	LY	P	C	P	I

Contd……..

Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type	Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type
51.	C	PR	Y	P	I	76.	LY	PR	Y	CY	I
52.	LY	R	C	CY	N	77.	Y	R	Y	CY	I
53.	Y	P	Y	CY	I	78.	LY	P	Y	CY	N
54.	LY	R	Y	P	F	79.	LY	R	Y	CY	N
55.	Y	P	C	P	N	80.	Y	PR	Y	CY	I
56.	LY	PR	Y	CY	I	81.	Y	P	C	CY	F
57.	Y	P	Y	CY	N	82.	LY	R	Y	CY	N
58.	LY	PR	Y	CY	I	83.	LY	P	C	P	I
59.	LY	P	C	CY	F	84.	Y	PR	Y	CY	I
60.	C	R	Y	P	N	85.	LY	R	Y	CY	F
61.	LY	PR	Y	CY	I	86.	Y	P	C	CY	N
62.	LY	P	C	P	I	87.	LY	PR	Y	P	I
63.	Y	PR	Y	CY	F	88.	LY	P	Y	CY	F
64.	C	R	Y	CY	I	89.	LY	PR	C	CY	N
65.	LY	PR	Y	CY	N	90.	Y	R	Y	P	I
66.	C	PR	Y	CY	F	91.	LY	P	Y	CY	F
67.	LY	P	C	CY	I	92.	LY	PR	C	CY	N
68.	LY	R	Y	CY	N	93.	C	PR	Y	P	I
69.	LY	P	Y	CY	I	94.	LY	R	C	CY	I
70.	C	PR	Y	CY	I	95.	C	P	Y	CY	N
71.	LY	P	C	CY	N	96.	LY	PR	Y	P	I
72.	C	R	Y	CY	F	97.	LY	R	C	CY	I
73.	LY	P	Y	CY	I	98.	C	P	Y	CY	I
74.	LY	P	Y	CY	N	99.	LY	PR	C	P	I
75.	Y	P	C	CY	F	100.	LY	R	Y	CY	I

Contd……..

Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type	Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type
101.	LY	PR	Y	CY	F	126.	Y	PR	C	CY	I
102.	C	PR	C	P	N	127.	LY	PR	Y	CY	N
103.	LY	R	Y	CY	I	128.	C	R	Y	CY	I
104.	Y	PR	C	CY	N	129.	LY	PR	Y	CY	N
105.	C	PR	Y	CY	I	130.	LY	R	Y	P	N
106.	LY	PR	Y	P	N	131.	Y	PR	Y	P	N
107.	Y	P	C	CY	F	132.	C	PR	Y	CY	I
108.	LY	PR	Y	CY	I	133.	LY	R	Y	CY	N
109.	Y	R	Y	CY	N	134.	LY	R	C	CY	I
110.	C	PR	C	P	I	135.	Y	PR	C	CY	F
111.	LY	PR	Y	CY	F	136.	LY	R	Y	CY	N
112.	Y	PR	Y	CY	N	137.	Y	PR	Y	CY	I
113.	C	PR	C	CY	N	138.	LY	P	Y	P	N
114.	LY	PR	Y	P	F	139.	Y	PR	Y	CY	I
115.	C	PR	Y	CY	F	140.	LY	R	C	CY	N
116.	LY	P	C	CY	N	141.	C	PR	Y	CY	I
117.	C	R	Y	CY	I	142.	LY	PR	C	CY	F
118.	LY	PR	Y	P	N	143.	LY	R	Y	P	N
119.	LY	PR	Y	CY	F	144.	Y	PR	Y	CY	F
120.	Y	PR	C	CY	N	145.	C	R	Y	CY	F
121.	LY	PR	C	CY	I	146.	LY	P	C	CY	I
122.	C	P	Y	P	F	147.	C	PR	Y	CY	N
123.	LY	PR	Y	CY	I	148.	LY	R	Y	CY	N
124.	C	R	Y	CY	N	149.	Y	PR	C	P	F
125.	LY	PR	Y	CY	I	150.	LY	PR	Y	CY	N

Contd……..

Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type	Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type
151.	LY	P	C	CY	I	176.	LY	R	Y	CY	F
152.	C	PR	Y	CY	F	177.	LY	PR	C	CY	I
153.	LY	R	C	CY	I	178.	Y	PR	Y	CY	N
154.	C	PR	Y	CY	F	179.	Y	R	Y	CY	I
155.	LY	R	C	P	F	180.	LY	PR	Y	CY	F
156.	LY	P	Y	CY	N	181.	C	PR	Y	CY	I
157.	C	PR	Y	CY	I	182.	LY	R	C	P	F
158.	LY	P	Y	CY	N	183.	Y	PR	Y	CY	I
159.	Y	PR	Y	CY	I	184.	LY	R	Y	CY	F
160.	Y	R	C	CY	I	185.	LY	PR	C	CY	I
161.	Y	R	Y	CY	I	186.	Y	PR	Y	CY	N
162.	LY	PR	Y	CY	I	187.	LY	R	Y	P	I
163.	Y	R	C	CY	I	188.	Y	P	Y	CY	F
164.	LY	PR	Y	CY	N	189.	LY	PR	C	CY	I
165.	Y	R	C	CY	F	190.	LY	R	Y	CY	N
166.	LY	PR	Y	P	I	191.	LY	PR	Y	CY	I
167.	LY	PR	Y	CY	N	192.	LY	PR	C	CY	I
168.	Y	R	Y	CY	I	193.	C	R	Y	CY	F
169.	LY	PR	C	CY	I	194.	LY	PR	Y	CY	I
170.	Y	R	Y	P	N	195.	LY	P	Y	P	N
171.	LY	PR	Y	CY	I	196.	C	PR	Y	CY	I
172.	LY	PR	CV	CY	F	197.	LY	PR	Y	CY	I
173.	Y	R	Y	CY	N	198.	LY	P	C	P	F
174.	LY	PR	C	CY	I	199.	Y	PR	Y	CY	I
175.	Y	PR	Y	P	N	200.	LY	R	Y	CY	N

Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type	Plant No.	Petal colour	Petal spot colour	Anther colour	Filament colour	Bract type
201.	LY	PR	C	CY	I	210.	LY	R	Y	CY	F
202.	Y	PR	C	CY	I	211.	Y	PR	C	CY	N
203.	LY	P	Y	CY	N	212.	LY	PR	Y	P	I
204.	Y	R	Y	CY	F	213.	Y	R	C	CY	I
205.	LY	PR	C	CY	I	214.	LY	PR	Y	CY	I
206.	C	PR	Y	P	I	215.	Y	R	C	CY	N
207.	Y	PR	Y	CY	N	216.	LY	PR	Y	P	I
208.	LY	R	C	CY	I	217.	Y	R	Y	CY	I
209.	Y	PR	Y	CY	F	218.	LY	R	C	CY	F

LY – Light yellow
P – Purple
C – Cream
N – Normal
Y – Yellow
F- Frego
PR – Purplish red
I – Intermediate
R – Red

Appendix III: Yield and yield contributing characters of F_2 population

Plant No.	No. of bolls	Seed cotton yield	Boll weight	Seed index	Plant No.	No. of bolls	Seed cotton yield	Boll weight	Seed index
1	4.00	3.00	0.75	7.50	41	5.00	3.00	0.60	7.10
2	11.00	11.00	1.00	7.30	42	41.00	59.00	1.44	7.30
3	106.00	88.00	0.83	7.50	43	60.00	41.00	0.68	7.10
4	70.00	77.00	1.10	7.60	44	9.00	8.00	0.89	7.20
5	4.00	3.00	0.75	7.30	45	237.00	269.00	1.14	7.30
6	32.00	29.00	0.91	7.10	46	3.00	1.00	0.33	7.50
7	15.00	19.00	1.27	7.50	47	12.00	9.00	0.75	7.10
8	17.00	14.00	0.82	7.40	48	6.00	9.00	1.50	7.50
9	35.00	29.00	0.83	6.90	49	22.00	11.00	0.50	6.90
10	11.00	8.00	0.73	7.40	50	58.00	53.00	0.91	7.10
11	10.00	9.00	0.90	7.50	51	20.00	24.00	1.20	7.40
12	10.00	9.00	0.90	7.40	52	11.00	7.00	0.64	6.50
13	6.00	7.00	1.17	7.60	53	14.00	9.00	0.64	6.80
14	51.00	62.00	1.22	7.50	54	33.00	25.00	0.76	7.10
15	49.00	54.00	1.10	7.20	55	8.00	7.00	0.88	7.20
16	98.00	132.00	1.35	7.20	56	7.00	7.00	1.00	7.30
17	87.00	92.00	1.06	7.20	57	23.00	24.00	1.04	7.30
18	4.00	3.00	0.75	7.40	58	9.00	4.00	0.44	7.10
19	20.00	27.00	1.35	7.30	59	41.00	26.00	0.63	7.40
20	5.00	2.00	0.40	7.10	60	27.00	21.00	0.78	7.20
21	25.00	13.00	0.52	7.50	61	17.00	17.00	1.00	7.20
22	6.00	2.00	0.33	6.90	62	13.00	14.00	1.08	7.20
23	13.00	13.00	1.00	6.50	63	3.00	2.00	0.67	7.30
24	4.00	4.00	1.00	6.80	64	8.00	6.00	0.75	7.10
25	2.00	1.00	0.50	7.10	65	46.00	58.00	1.26	7.10
26	159.00	188.00	1.18	7.40	66	17.00	8.00	0.47	6.90
27	2.00	4.00	2.00	7.10	67	4.00	3.00	0.75	6.90
28	6.00	30.00	5.00	7.20	68	7.00	6.00	0.86	7.20
29	2.00	1.00	0.50	7.50	69	25.00	17.00	0.68	7.30
30	15.00	17.00	1.13	6.90	70	25.00	24.00	0.96	7.30
31	3.00	3.00	1.00	7.40	71	25.00	26.00	1.04	7.10
32	22.00	23.00	1.05	7.10	72	19.00	16.00	0.84	7.10
33	57.00	47.00	0.82	7.20	73	17.00	10.00	0.59	7.50
34	80.00	84.00	1.05	7.40	74	64.00	67.00	1.05	7.60
35	39.00	43.00	1.10	7.10	75	14.00	9.00	0.64	7.10
36	54.00	59.00	1.09	7.80	76	6.00	5.00	0.83	7.30
37	74.00	67.00	0.91	7.50	77	3.00	2.00	0.67	7.60
38	56.00	32.00	0.57	7.20	78	50.00	58.00	1.16	7.40
39	7.00	5.00	0.71	7.30	79	85.00	57.00	0.67	7.10
40	26.00	22.00	0.85	7.10	80	5.00	8.00	1.60	7.30

Contd….

Plant No.	No. of bolls	Seed cotton yield	Boll weight	Seed index	Plant No.	No. of bolls	Seed cotton yield	Boll weight	Seed index
81	9.00	11.00	1.22	7.20	119	12.00	11.00	0.92	7.10
82	32.00	32.00	1.00	7.40	120	5.00	2.00	0.40	7.20
83	2.00	2.00	1.00	6.80	121	20.00	23.00	1.15	7.50
84	10.00	15.00	1.50	6.90	122	7.00	15.00	2.14	6.80
85	10.00	8.00	0.80	7.10	123	23.00	32.00	1.39	6.70
86	6.00	6.00	1.00	7.20	124	4.00	1.00	2.00	7.20
87	10.00	5.00	0.50	7.20	125	6.00	2.00	1.00	7.30
88	48.00	56.00	1.17	7.30	126	5.00	5.00	1.00	7.00
89	39.00	39.00	1.00	6.90	127	14.00	16.00	1.14	6.50
90	42.00	37.00	0.88	7.10	128	42.00	63.00	1.50	6.20
91	4.00	3.00	0.75	7.20	129	115.00	194.00	1.69	7.00
92	10.00	9.00	0.90	7.20	130	66.00	99.00	1.50	7.30
93	40.00	34.00	0.85	7.10	131	118.00	184.00	1.56	7.20
94	10.00	7.00	0.70	7.50	132	53.00	69.00	1.30	6.90
95	40.00	39.00	0.98	7.30	133	78.00	107.00	1.37	7.50
96	3.00	5.00	1.67	7.30	134	61.00	83.00	1.36	7.10
97	17.00	18.00	1.06	7.20	135	36.00	58.00	1.61	7.20
98	4.00	3.00	0.75	7.20	136	54.00	82.00	1.52	7.10
99	9.00	9.00	1.00	7.20	137	19.00	24.00	1.26	7.00
100	49.00	69.00	1.41	7.10	138	44.00	69.00	1.57	7.60
101	3.00	1.00	0.33	7.20	139	25.00	41.00	1.64	6.80
102	16.00	18.00	1.13	7.30	140	7.00	15.00	2.14	6.50
103	16.00	13.00	0.81	7.50	141	11.00	23.00	2.09	6.40
104	2.00	1.00	0.50	7.40	142	19.00	33.00	1.74	7.10
105	2.00	1.00	0.50	7.20	143	64.00	102.00	1.59	7.20
106	35.00	31.00	0.89	7.20	144	20.00	24.00	1.20	7.10
107	5.00	3.00	0.60	7.10	145	109.00	139.00	1.28	7.20
108	15.00	9.00	0.60	7.10	146	8.00	11.00	1.38	7.20
109	5.00	2.00	0.40	7.60	147	2.00	2.00	1.00	6.90
110	9.00	8.00	0.89	7.50	148	10.00	13.00	1.30	7.30
111	6.00	5.00	0.83	7.40	149	21.00	26.00	1.24	6.80
112	8.00	6.00	0.75	7.10	150	15.00	17.00	1.13	6.50
113	12.00	11.00	0.92	7.20	151	21.00	27.00	1.29	6.20
114	2.00	1.00	0.50	6.70	152	12.00	9.00	0.75	7.10
115	10.00	7.00	0.70	7.10	153	19.00	21.00	1.11	6.40
116	11.00	8.00	0.73	6.90	154	25.00	34.00	1.36	7.40
117	10.00	6.00	0.60	6.90	155	53.00	71.00	1.34	7.20
118	13.00	10.00	0.77	7.40	156	37.00	39.00	1.05	6.80

Contd….

Plant No.	No. of bolls	Seed cotton yield	Boll weight	Seed index	Plant No.	No. of bolls	Seed cotton yield	Boll weight	Seed index
157	3.00	4.00	1.33	7.40	195	10.00	23.00	2.30	6.50
158	69.00	132.00	1.91	7.20	196	28.00	30.00	1.07	7.10
159	9.00	10.00	1.11	7.10	197	46.00	60.00	1.30	7.20
160	29.00	44.00	1.52	7.30	198	28.00	39.00	1.39	7.10
161	11.00	15.00	1.36	7.60	199	29.00	34.00	1.17	7.10
162	39.00	50.00	1.28	7.10	200	29.00	28.00	0.97	7.30
163	16.00	23.00	1.44	7.20	201	19.00	25.00	1.32	7.20
164	14.00	16.00	1.14	7.30	202	18.00	26.00	1.44	7.10
165	9.00	13.00	1.44	7.20	203	88.00	162.00	1.84	6.90
166	2.00	5.00	2.50	7.10	204	55.00	71.00	1.29	6.90
167	38.00	48.00	1.26	7.60	205	18.00	26.00	1.44	7.60
168	48.00	61.00	1.27	7.40	206	24.00	33.00	1.38	7.50
169	31.00	44.00	1.42	6.90	207	36.00	43.00	1.19	7.40
170	2.00	1.00	0.50	7.20	208	11.00	17.00	1.55	7.10
171	28.00	43.00	1.54	7.10	209	14.00	17.00	1.21	6.90
172	24.00	32.00	1.33	7.30	210	29.00	38.00	1.31	7.20
173	36.00	39.00	1.08	7.10	211	37.00	47.00	1.27	7.20
174	22.00	31.00	1.41	6.80	212	26.00	32.00	1.23	6.90
175	3.00	2.00	0.67	6.60	213	28.00	33.00	1.18	7.40
176	32.00	46.00	1.44	7.20	214	33.00	42.00	1.27	7.60
177	5.00	3.00	0.60	7.20	215	24.00	25.00	1.04	7.30
178	8.00	7.00	0.88	7.10	216	17.00	21.00	1.24	7.20
179	5.00	4.00	0.80	6.80	217	22.00	28.00	1.27	7.20
180	13.00	14.00	1.08	6.40	218	20.00	21.00	1.05	7.10
181	67.00	78.00	1.16	7.10					
182	17.00	31.00	1.82	6.40					
183	37.00	42.00	1.14	7.20					
184	6.00	7.00	1.17	6.30					
185	3.00	4.00	1.33	7.10					
186	20.00	30.00	1.50	7.20					
187	8.00	11.00	1.38	7.10					
188	17.00	30.00	1.76	7.40					
189	5.00	3.00	0.60	7.20					
190	16.00	13.00	0.81	7.00					
191	22.00	30.00	1.36	6.90					
192	24.00	20.00	0.83	7.10					
193	3.00	3.00	1.00	6.80					
194	62.00	81.00	1.31	7.30					

Appendix IV: Halo length of F_2 plants

Plant No.	Halo length	Plant No.	Halo length	Plant No.	Halo length	Plant No.	Halo length	Plant No.	Halo length
1	17.89	52	20.00	103	22.89	154	20.11	205	21.67
2	19.56	53	19.33	104	17.11	155	20.56	206	19.67
3	17.56	54	20.44	105	18.56	156	20.56	207	19.22
4	18.11	55	20.00	106	19.11	157	18.11	208	20.11
5	19.11	56	18.22	107	13.56	158	17.89	209	18.89
6	18.67	57	24.44	108	16.44	159	16.22	210	19.78
7	20.67	58	20.67	109	19.33	160	18.67	211	20.67
8	22.44	59	18.11	110	21.22	161	17.11	212	18.78
9	18.22	60	22.78	111	14.44	162	20.33	213	19.78
10	17.67	61	22.22	112	21.33	163	18.56	214	17.78
11	21.89	62	21.11	113	17.44	164	17.44	215	18.22
12	17.67	63	22.33	114	19.33	165	16.56	216	20.00
13	19.78	64	20.56	115	15.33	166	20.22	217	19.22
14	18.22	65	19.11	116	21.33	167	20.11	218	18.33
15	20.89	66	20.00	117	19.11	168	19.44	Mean	19.58
16	16.22	67	20.00	118	20.44	169	14.89		
17	16.44	68	18.33	119	19.56	170	19.11		
18	16.22	69	20.11	120	19.78	171	19.67		
19	18.56	70	21.11	121	17.67	172	21.44		
20	17.67	71	20.11	122	17.00	173	18.78		
21	17.78	72	21.78	123	19.67	174	18.11		
22	17.22	73	19.44	124	15.44	175	20.89		
23	25.44	74	20.33	125	20.11	176	21.78		
24	19.89	75	20.89	126	17.67	177	22.00		
25	22.33	76	18.67	127	21.22	178	17.00		
26	17.22	77	18.33	128	20.22	179	18.89		
27	23.56	78	21.44	129	20.67	180	18.67		
28	18.44	79	16.56	130	18.89	181	20.33		
29	15.22	80	18.89	131	19.00	182	26.00		
30	19.67	81	19.00	132	23.11	183	24.44		
31	16.33	82	19.56	133	20.89	184	18.11		
32	24.11	83	22.22	134	20.44	185	19.89		
33	17.44	84	21.89	135	20.44	186	16.44		
34	19.00	85	22.44	136	19.22	187	22.00		
35	22.11	86	20.67	137	20.11	188	18.89		
36	20.67	87	18.56	138	19.89	189	19.33		
37	20.56	88	21.00	139	18.33	190	19.78		
38	19.56	89	20.33	140	21.33	191	22.56		
39	18.33	90	21.78	141	20.11	192	20.56		
40	19.33	91	20.78	142	18.67	193	21.33		
41	16.67	92	20.00	143	20.11	194	23.67		
42	20.44	93	21.56	144	19.44	195	26.33		
43	19.33	94	18.00	145	19.44	196	18.11		
44	23.44	95	21.67	146	17.56	197	20.22		
45	19.33	96	18.00	147	18.67	198	18.67		
46	18.33	97	19.67	148	19.67	199	16.44		
47	19.33	98	18.67	149	19.22	200	17.44		
48	17.89	99	21.67	150	17.56	201	22.00		
49	21.33	100	19.56	151	19.22	202	21.22		
50	19.11	101	18.78	152	18.33	203	20.78		
51	20.00	102	19.00	153	19.22	204	21.00		

Appendix V: List of polymorphic primers and their per cent polymorphism in DDhc-11 and *G. anomalum*

Sl. No.	Primers	Total No. of bands	No. of polymorphic bands	Per cent polymorphism
1.	OPA-9	6	2	33.3
2.	OPC-1	6	1	16.6
3.	OPC1-2	5	4	80.0
4.	OPC-9	7	2	28.5
5.	OPP-7	4	1	25.0
6.	OPB-11	8	6	75.0
7.	OPC-20	5	2	40.0
8.	OPY-18	6	4	66.6
9.	OPP-12	10	8	80.0
10.	OPP-6	7	5	71.4
11.	OPZ-19	5	4	80.0
12.	OPP-13	4	3	75.0
13.	OPP-9	9	2	22.2
14.	OPY-11	7	5	71.4
15.	OPY-9	8	2	25.0
16.	OPC-11	5	3	60.0
17.	OPY-20	6	2	33.3
18.	OPB-01	5	3	60.0
19.	OPZ-13	8	1	12.5
20.	OPY-14	4	2	50.0
21.	OPA-2	6	4	66.6
	Total	**131**	**66**	**51.0**
	No of bands per primer	**6.23**	**3.14**	

STUDIES ON GENETIC INTROGRESSION IN COTTON THROUGH INTERSPECIFIC HYBRIDIZATION

Ch. MADAN MOHAN **2005** **Dr. P. M. SALIMATH**
Major Advisor

ABSTRACT

The present investigation was carried out at Agricultural Research Station, Dharwad Farm during *kharif* 2004-05 to study the genetic introgression in an interspecific cross between *G. herbaceum* var. DDhc-11 and *G. anomalum*. *G. herbaceum* var. DDhc-11, which is cultivated diploid variety is having some constraints with respect to fibre properties in order to improve fibre properties of DDhc-11, *G. anomalum* a lintless diploid wild relative was used as male parent. The present study was aimed to develop recombinant plants with superior fibre properties.

Distinguishable morphological qualitative characters such as petal colour, petal spot colour, anther colour, filament colour and bract type were studied. All these characters fitted in to monohybrid ratios. Plants, which possessed higher halo length and sufficient lint were subjected for detailed study. The mean number of bolls per plant and GOT of selected F_2 plants showed significant superiority and inferiority, respectively over the female parent DDhc-11. Whereas, seed cotton yield and seed index showed non-significant superiority and boll weight and lint index showed non-significant inferiority.

Twenty two plants were analysed for fibre properties for characters like fibre strength, fibre fineness and elongation percentage F_2 plants showed had shown significant superiority over DDhc-11. For fibre length, uniformity ratio and maturity ratio numerical superiority was observed.

Out of 5.4 fragments per primer amplified, 3.58 fragments per primer were polymorphic. The cluster diagram and dice similarity coefficients indicated 60 to 82 per cent similarity between selected F_2 plants and the parents. The *G. anomalum* formed separate cluster. The molecular markers indicated an average of 76.4 per cent of specific bands from DDhc-11 and only 42.3 per cent of specific bands from *G. anomalum*.

Based on fibre strength values, 22 plants were categorized into nine groups representing the fibre strength from 17 to 25 g per tex. OPZ-19 amplified a 300 bp amplicon, which was present only in groups representing 23, 24 and 25 g per tex along with P_2 and F_1, but was absent in other groups indicating its association with fibre strength.