Exotic Germplasm Introgression Effects on Agronomic Traits and Fiber Properties of Upland Cotton

by

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Abstract

Upland cotton (*Gossypium hirsutum* L.) is an economically important natural fiber crop in the world. Along with fiber, it is also valued for its oil and protein portion of the seed. Upland cotton is facing a risk associated with a narrow genetic base. The utilization of closely related parents and many reselections within elite cultivars has led to a narrow genetic base. Cotton cultivars with increased genetic diversity can offer plasticity to be able to respond to stressful environments. Our research is focused on the objective of determining the effect of exotic germplasm on agronomic traits and fiber properties of adapted cotton cultivars. Two plant introductions were systematically crossed with four cultivars to derive five groups of lines with various levels of exotic introgression. Experimental materials were tested in a randomized complete block design and analyzed using statistical analysis software.

Results revealed highly significant effects due to population, exotic percentage, and population × exotic percentage interactions for all the agronomic and fiber traits except for boll number. An increase in exotic percentage significantly lowered agronomic and fiber properties, however the magnitude of effect varied with populations and exotic percentage levels. Among agronomic traits seed cotton yield, lint seed^{-1}, lint percentage and lint yield were the most affected, whereas days to first flower, bolls plant^{-1}, boll size, seeds boll^{-1} and lodging were least affected with an increase in exotic percentage. In most populations an increase in exotic percentage did not show any significant difference up to 25 percent exotic germplasm introgression for all the agronomic traits except for lint mass seed^{-1}. Among fiber properties,

micronaire was the most affected trait, whereas fiber elongation and short fiber content were the least affected traits with an increase in exotic introgression. In almost all cases where the exotic parent was better or equal to the performance of adapted cultivars, an increase in exotic percentage up to 25 percent did not show a significant difference in fiber properties, which suggests that using exotic parents which are similar or better in fiber properties could expand genetic base of adapted cultivars without altering fiber properties.

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Table of Contents

List of Tables

List of Figures

List of Abbreviations

EP	Exotic percentage
POP	Population
$F_{2:4}$	Second filial generation where selection was performed and maintained further
BC_1	Backcross one generation
SE	Standard Error
ANOVA	Analysis of variance
COV	Covariance
PI	Plant Introduction
LS	Least square

1. Literature review

Genetic diversity is defined as the total heritable variation present within a plant species (Brown, 1983). Genetic diversity occurs mainly due to genetic changes that may arise due to differences in DNA sequence, biochemical compounds, or physiological changes due to stress and morphological adaptations. Genetic diversity in plant populations may change due to temporal and spatial structures (Loveless and Hamrick, 1984). In a broad view, genetic diversity may depend on many factors such as evolution, breeding systems, ecological, geological and often by anthropic factors (Ramanatha Rao and Hodgkin, 2002). Genetic diversity can be looked on two different levels; parental and regional (NAS, 1972). Parental diversity is the amount of variability present among the parents. It is a key tool in hybrid development since some degree of parental diversity is important for basic expression of hybrid vigor. Regional diversity is determined by the number of different cultivars planted in a region. Lack of diversity at either or both parental and regional level may predispose crops to serious pest and disease outbreaks.

Importance of genetic diversity:

Genetic diversity is an invaluable resource for selection and improvement in crop cultivar development. Genetic diversity is a key contributor to genetic variation for any trait, and as such is a major factor in achieving genetic gain. The success of a breeder in a highly domesticated, autogamous crop depends mainly on identifying superior lines to use as parents, so having diverse genetic pool offers flexibility for selection. The importance of genetic diversity was

emphasized many times in the literature (Duvick, 1984; Meredith, Jr., 1991; Wallace et al., 2009). Genetic diversity offers plasticity to be able to respond to stressful environments.

Genetic vulnerability:

Lack of genetic variability may lead to a serious phenomenon called genetic vulnerability (Acquaah, 2007). The possible threat due to lack of genetic variability in several crops has been reported (Hammons, 1976; Walsh, 1981). Mainstream breeding programs primarily focus on developing high yielding, high quality and uniform cultivars in a short period of time, so the first choice of parents would be selections from a few available elite lines or the lines which were developed through repeated backcross or reselections in the past. This breeding practice over a period of time unintentionally narrows the genetic base of the crop species. When these genetically uniform varieties are cultivated on a larger scale over larger areas and coupled with certain predisposing conditions plants may become genetically vulnerable to pest and diseases resulting in disease outbreaks. There are numerous examples from the past where lack of genetic diversity led to disease outbreaks or epidemics. Some of the most disastrous diseases resulting from lack of genetic diversity occurred in past were late blight of potato (*Phytopthora infestans* (Mont.) de Bary), coffee rust (*Hemilea vastatrix*), brown spot of rice (*Helminthosporium oryzae* Breda de Haan), Panama wilt on banana (*Fusarium oxysporum* Schltdl. f. sp. *cubense* (E. F. Smith.) W. C. Snyder & H. N. Hansen), and Southern leaf blight on corn (*Helminthosporium maydis*) in the US.

The Irish famine in 1840's (caused by *Phytopthora infestans* (Mont.) de Bary) claimed more than one million lives and led one more million people to emigrate from Ireland to other countries due to the shortage of food (Smart and Fry, 2001). Genetic uniformity and monoculture potato cultivar Irish Lumper was the predisposing cause for this epidemic. Coffee rust caused by

Hemilea vastatrix severely affected Arabica coffee plantations during the last quarter of 19th century. Cultivars grown in Ceylon during that time period were all genetically similar as they were derived from a single source of *Coffea arabica* L. Because of this, the coffee crop failed completely (Rodrigues et al., 1975). The great Bengal famine in 1943 was caused by brown spot of rice (*Helminthosporium oryzae* Breda de Haan) resulted in an estimated death toll of two million people in India (Padmandabhan, 1973). Panama wilt in banana caused by *Fusarium oxysporum* Schltdl. f. sp. *cubense* (E. F. Smith.) W. C. Snyder & H. N. Hansen affected the banana industry severely during 1950's (Ploetz and Pegg, 1997). Large-scale cultivation of a single planting material 'Gros Michel' over a period of time and using infected rhizomes for propagation led to this outbreak. Genetic diversity received a special attention in the US as a result of southern corn leaf blight, which occurred in 1970. Before 1970, corn was usually considered a relatively healthy crop, but the epidemic in 1970 brought out the susceptibility, which came about largely due to the techniques for producing commercial hybrid seed. It started with the transfer of a male sterile cytoplasm called cms-T (Texas male sterile cytoplasm) to almost all inbred lines through repeated backcrossing for 6-8 generations. By 1970, 85 percent of hybrid seed corn produced contained this cms-T cytoplasm, which was hyper-susceptible to the pathogen *Helminthosporium maydis* leading to a devastating epidemic called Southern corn leaf blight. The official estimate of the loss incurred due to this epidemic was almost one billion dollars in the United States (Tatum, 1971; Ullstrup, 1972). It is also important to watch out for some diseases that fit into a category called "emerging diseases", which start off as minor diseases and may turn out to be epidemics and cause serious losses. One pathogen that is emerging as a possible threat to American and worldwide grape production is *Xylella fastidiosa*, the causal agent of Pierce's disease in grapes. Almost all popular cultivars of

European, American and French-American hybrid grape types are susceptible to this bacterium, but resistance was found in *Vitis* species types native to the Southeast US (Hopkins and Purcell, 2002). These are a few of many examples where genetic vulnerability coupled with disease outbreaks has led to serious losses and claimed many human lives. We should also be aware that there might be many disasters brewing right now because of the genetic vulnerability resulting due to genetic uniformity. Hence, it is very desirable to improve or broaden the genetic base of the current day crop cultivars.

Exotic germplasm:

Exotic germplasm refers to germplasm that does not have an immediate utility, unless selection pressure is applied for adaptation (Hallauer and Miranda, 1981; Holland, 2004). Since it is not subjected to extensive selection, much of the diversity is supposed to be preserved in its natural form. Exotic germplasm is a valuable resource of variability but much of its potential still remains underutilized. Complications like linkage drag, high level epistasis (nonlinear interaction with unlinked genes), beneficial QTLs showing significant opposite effect in different genetic backgrounds, restricted recombination, hybrid break down, requiring more time and effort, and other important drawbacks presents a difficult challenge for exotic introgression (Jiang et al., 2000; Paterson et al., 2004; Zhong et al., 2002). Chang and Seshu (1990) indicated that exotic germplasm also suffers constraints such as lack of adaptation to commercial production, poor agronomic characteristics, low crossability and sterility, taxonomical ambiguity and fertility relationships. However, with the assistance of modern techniques like tissue culture, marker assisted selection and biotechnological tools, some of these limitations can be overcome.

Utility of exotic germplasm:

Historically, exotic germplasm has been utilized as a source of resistance to pest, diseases, biotic and abiotic stresses in many crops over time (Hajjar and Hodgkin, 2007). Using exotic germplasm as a source for improving yield has not been particularly looked at due to the complex nature of yield as a trait. Schoener and Fehr (1979) indicated that exotic germplasm with good yield potential can be utilized in increasing variability without drastically reducing population performance in a soybean study. Exotic germplasm may not be a poor contributor for yield, but there is currently not enough understanding or information about the effects of exotic germplasm on yield (Holland, 2004). Using exotic germplasm for yield improvement in short term vs. long term improvement may also have a bearing on the successful utilization of these resources (Ininda et al., 1996). Vello et al. (1984) reported that the use of plant introductions might not be productive if used as experimental lines in short term improvement. Cox et al. (1984) indicated long-term recurrent selection studies are needed to determine the actual potential of plant introductions in improving yield. Schoener and Fehr (1979) proposed a similar kind of opinion that using plant introductions in short term yield improvement has no advantage. Some examples in which exotic germplasm played significant role are presented below.

In barley, resistance to soil-borne barley mild mosaic virus (BaMMV) was found in exotic germplasm sources of *H. vulgare* L and *H. spontaneum* Koch (Ordon et al., 1996). In wheat, multiple disease resistance and vigorous seedling emergence was found in PI 178383, and subsequently transferred to bread wheat. In rice, a gene for the semi-dwarf trait was instrumental in the Green Revolution. Wild abortive 'CMS-WA' for cytoplasmic male sterility and grassy stunt virus resistance in rice could be considered as an important contribution of exotic germplasm. And many other crops like corn, oat, sugarcane, potato etc have been the beneficiaries of improvement from exotic germplasm.

Cotton:

Cotton, the white gold, also known as king of fiber crops, is one of the most important commercial cash crops in the world. Cotton is among the first crops in which the rediscovered Mendelian principles were applied (Balls, 1907). Cotton has a predominant status among all the commercial crops providing cotton fiber for the textile industry. It is also valued for the protein and oil portion of the seed. The protein portion of the seed is mainly utilized in cattle feed and the oil portion as a vegetable oil for the food industry and in industrial usage such as lubricants. Despite severe competition from the synthetic fiber industry in recent years, cotton is still holding its commercial value as an important natural fiber crop in the textile industry.

Origin and evolution of cotton:

The genus *Gossypium* belongs to the family Malvaceae (Brubaker et al., 1999; Fryxell, 1968, 1992). It contains approximately 50 species out of which 45 are diploids and 5 are tetraploids (Fryxell, 1992). The origin of tetraploid *Gossypium* species dates back to one to two million years ago (Wendel, 1989), when hybridization happened between two diploid *Gossypium* species with two different genomes, one with an old world 'A' genome (relative of *G. herbaceum* L and *G. arboreum* L (2n=2x=26)) and the other with 'D' genome (relative of *G. raimondii* Ulbrich and *G. gossopoides* L (2n=2x=26)) (Beasley, 1940a, 1942; Wendel et al., 1992). This hybridization was followed by chromosome doubling resulted in an abrupt speciation (Fryxell, 1965) of an allotetraploid *Gossypium* spp with 'AADD' genome (2n=4x=52) (Beasley, 1940b; Paterson et al., 2004; Wendel and Cronn, 2002). The 'AADD' tetraploid emerged into the new world and gave rise to five tetraploid *Gossypium* species including the

world's most important commercially cultivated cottons namely, upland cotton (*G. hirsutum* L) and Egyptian cotton (*G. barbadense* L) (Wendel, 1989).

Genetic potential and its utilization in cotton:

The genetic potential in cotton is reported to be vastly large (Bowman et al., 1996; Meyer, 1974; Percival and Kohel, 1990). Stewart (1994) categorized *Gossypium* species into three germplasm pools (primary, secondary and tertiary) based on the affinity and ease of crossing. According to his classification, primary germplasm pool includes species that are easily crossable and produce highly fertile hybrids; all tetraploid species belong to this pool. Secondary and tertiary germplasm pools include species that require special manipulation techniques for successful crosses. Species in these pools generate low fertility hybrids. At present, the primary germplasm pool consists of five tetraploid species, the secondary germplasm pool consists of 20 species and the tertiary germplasm pool consists of 25 species (Campbell et al., 2010). Among the five tetraploid species, upland cotton (*G. hirsutum* L) and pima cotton (*G. barbadense* L) are the most widely cultivated. Currently, the most widely grown industrial cotton is upland cotton (*G. hirsutum*), which accounts for more than 90 percent of world production area (Campbell et al., 2010; Wendel et al., 1992).

The traits we see today in cotton are the result of repeated human selection and domestication. Much of the genetic variability preserved once in the form of wild relatives or feral relatives has been depleted because of improper attention and lack of sufficient expertise to efficiently utilize them in the cultivar development. Campbell et al. (2010) indicated that there are number of germplasm collections which are unreported, underrepresented, and poorly maintained due to lack of information which may soon become extinct if neglected. Campbell et

al. (2010) and Wallace et al. (2009) covered the status of global cotton germplasm resources and the US cotton germplasm collection in depth.

Bowman (1999) provided three contributing factors for the improper utilization of genetic material available in cotton. 1) The main breeding method followed over years (Reselection); Breeders utilized established cultivars as a source in their breeding program and reselections from the same source may not necessarily bring any new variability except capitalizing on the residual heterozygosity. 2) Repeated use of same parents leading to genetic uniformity; Bowman et al. (1996) reported that Stoneville relatives appeared 383 times in the pedigrees of 216 cultivars released in between 1970 to 1990. 3) Reluctance of breeders to use unadapted germplasm: Van Esbroeck and Bowman (1998) reported that only four upland cotton lines occurred repeatedly in the pedigrees of upland cultivars even though a number of germplasm lines has been released since 1972.

Paterson et al. (2004) described three bottlenecks, which might have played an important role in narrowing the genetic base of cotton. 1. The monophyletic origin of tetraploid cottons; Out of eight diploid cotton species only two contributed for the formation of cultivated tetraploid cottons. 2. Selecting very few wild cotton genotypes for domestication. 3. Human sampling of tetraploid cotton genotypes from centers of diversity. A fourth bottleneck could be added because of the recent deployment of transgenes in only few cultivars (Gingle et al., 2006). Currently, almost all the contemporary commercial transgenic cotton cultivars are all derived by backcrossing to a transgenic non-recurrent donor parent, Coker 312 (Wallace et al., 2009). Bowman et al. (2003) reported that a shift in cotton production to transgenics has exacerbated the narrowing of the genetic base of modern cotton cultivars and reduced the potential for genetic gain of lint yield. Backcrossing to transgenics has also reduced genetic gain for fiber strength

(Bowman and Gutierrez, 2003). The incorporation of *Bt* genes (Jenkins et al., 1997) and glyphosate resistance genes (Johnson, 1996) greatly altered cotton cultivation trends. By the year 2000, nearly 54 percent of the US cotton market and 72 percent of the US acreage was occupied by transgenic cultivars (Bowman et al., 2003).

A survey was conducted with both public and private breeders in the US to understand the type of breeding methods used. Results indicated that public breeders concentrate approximately 50 percent on cultivar development and approximately 50 percent on germplasm enhancement. Private breeders on the other hand spend almost 100 percent of their effort in cultivar development with 65 percent using conventional breeding methods and 35 percent on using transgenic breeding methods (Bowman, 2000). This indicates that the effort spent on germplasm enhancement is significantly less than on cultivar development. In addition, curtailing free exchange of genetic material between public and private breeding programs because of patenting genetic material is also a limiting factor in expanding genetic base of cotton.

Studies on genetic base in cotton cultivars:

The current cotton genetic pool is narrow and lacking variability for economically important agronomic and fiber traits (Bowman, 1999; Bowman et al., 2006; Paterson et al., 2004). Unless new genetic diversity is brought in, it may be difficult to maintain genetic gains. The genetic base of modern cultivars has been studied extensively using both conventional pedigree based methods and modern biotechnological molecular marker based methods. Pedigree based methods used coefficient of parentages and field uniformity to estimate genetic uniformity, whereas molecular marker based methods used DNA based markers and enzyme based markers to estimate genetic uniformity. Although the information from pedigree studies is

valuable, the diversity estimates from these studies should be used carefully. Van Becelaere et al. (2005) indicated that the genetic similarity estimates based on COP (r_p) may not be a best measure to determine the genetic resemblance of cultivars; many of the contributing factors such as selection, mutation and genetic drift are not taken into consideration in calculating COP (r_p). Van Esbroeck et al. (1999) indicated that pedigree-based studies must have overestimated genetic distances because of the relatedness in the ancestral lines. Some of the studies using both methods are described in the following review.

Pedigree based studies used coefficient of parentage (r_p) as the measure of estimating genetic diversity, which is calculated using allelic identity by descent. It is the probability of two genotypes having the same allele at a random locus (Bowman et al., 1997). A value closer to '0' indicates it is genetically very diverse or no relationship, whereas a value closer to '1' indicates it is highly uniform or identical.

Genetic diversity was estimated for 126 upland cotton cultivars released between 1980 and 1990 using the coefficient of parentage (r_p) values published in Bowman et al. (1997). Cluster analysis resulted in 12 distinct gene pools and the calculated mean r_p value was 0.07, which indicated substantial diversity. However, the data suggested a downward trend in the genetic base of upland cotton because of the repetitive use of same parents and the increase in proprietary nature of germplasm (Bowman et al., 1996; May et al., 1995). Similar study using the pedigree information (Calhoun et al., 1994) of the 260 upland cotton varieties released in the USA from 1970 to 1995 revealed that an average of 17 (range = 8-46) cultivars accounted for more than 97 percent of cotton hectarage within a region.

Van Esbroeck et al. (1998) calculated the field uniformity values (r_f) and found that that field uniformity rested around 0.30 (r_f) in all the four cotton-growing regions in the US from

1970 to 1995. The mean regional coefficient of parentage (r_p) ranged from 0.12 to 0.15 between 1970 to1990, and increased to 0.20 in 1995. In spite of an increase in r_p values, the field uniformity coefficient stayed about the same because of the increase in the availability of new cultivars and the decrease in the proportion of planting single popular cultivars. All these studies suggest the availability of genetic diversity, but only few germplasm lines have been used in the development of commercial varieties. Pedigrees of upland cotton cultivars released during 1970 to 1995 contained a large number of reselections as their parental material (Bowman et al., 1996, 2003; May et al., 1995; Van Esbroeck et al., 1998).

The average coefficients of parentage in some other crops reported were 0.13 for 258 soybean cultivars released between 1947 and 1988 in North America (Gizlice et al., 1993), 0.17 for 122 elite soybean lines released in the southern and northern US (Sneller, 1994), 0.32 and 0.21 for runner and Virginia types respectively, in peanut cultivars released between 1969 and 1988 (Knauft and Gorbet, 1989), 0.09 and 0.07 for 2 and 6 rowed respectively, spring barley cultivars released between 1970 and 1990 (Martin et al., 1991), 0.19 for soft red wheat and 0.26 for hard red wheat cultivars (Murphy et al., 1986), 0.15 to 0.42 in oat cultivars released between 1941 to 1980, and finally 0.07 in 260 cotton cultivars released between 1970 and 1990 (Bowman et al., 1996). By considering the coefficients of parentage in all these other crops it would be fair to say that the genetic variability present in the cotton cultivars released during 1970 to 1990 was relatively better. However, it is to be noted that these coefficients of parentage in other crops were from variable time periods.

Wendel et al. (1992) conducted a study to assess the levels and patterns of genetic variation in upland cotton using Isozyme analysis. Results revealed that the genetic diversity in upland cotton had slightly higher percentage of polymorphic loci (60 percent compared to 49

percent in an "average" crop species) and slightly more polymorphic loci (2.30 alleles per a locus compared to 2.15), but recorded slightly lesser genetic diversity than average total diversity of other crop species. It was also observed that genetic diversity in *G. hirsutum* is better than all other three cultivated cotton species. A similar study using RFLP analysis also exhibited lower genetic diversity in *G. hirsutum* compared to other crop species (Brubaker and Wendel, 1994). Another study was conducted using 23 elite commercial cotton cultivars (which occupied 37 percent of acreage in the USA) along with a check TM-1 to evaluate the genetic diversity using 88 SSR primers. Genetic similarity coefficients ranged from 0.694 to 0.936 with an average of 0.772. Zhang et al. (2005) indicated that the genetic similarity coefficient value of 0.772 suggests that there is sufficient amount of genetic diversity present in those 23 elite commercial cotton cultivars studied (Zhang et al., 2005). Similar research studies conducted to estimate genetic diversity in cotton using various molecular markers such as RFLP (Brubaker and Wendel, 1994;Van Becelaere et al., 2005), AFLPs (Abdalla et al., 2001; Iqbal et al., 2001), rDNA (Pillay and Myers, 1999), SSRs (Gutierrez et al., 2002) and RAPDs (Linos et al., 2002) indicated low levels of genetic diversity in cotton.

The above studies clearly support the view that upland cotton is resting on a very narrow genetic base and is facing a risk associated with it (Campbell et al., 2009; May et al., 1995; McCarty et al., 2007; Paterson et al., 2004; Zeng et al., 2011). These above studies also explain limited improvement and or stagnant yield and fiber properties since 1980's (Chaudry, 1997; Felker, 2001; Fok, 1998; Helms, 2000; Lewis, 2001; Meredith, Jr., 1991, 2000, 2005)

Genetic resources and ways to improve genetic diversity:

The importance of genetic resources should be well understood and documented in order for it to be conserved, utilized and handled efficiently (Campbell et al., 2010). There is no better

way than collecting and preserving germplasm resources which may otherwise be lost if proper care is not taken. The National Plant Germplasm System is an organization that collects and preserves the resources. The recent germplasm collection trips by the NPGS have added 12 new *Gossypium* species to germplasm collection. The germplasm resources available at this center are collected by spending lot of effort and time. At present, NPGS holds 9332 accessions representing 49 *Gossypium* species from 74 different countries (Wallace et al., 2009). Utilizing these resources in the breeding program at some level or degree could be a better approach of improving genetic variability. Introgressing novel genes from rich sources of diversity like landraces, obsolete cultivars, plant introductions, wild races and feral species may alleviate the problem of genetic vulnerability. When we introduce variability, we are not only introducing favorable genes from the formerly mentioned sources, but also potential problems such as linkage drag, hybrid breakdown and sterility. In order to successfully improve the genetic base we should know the basic mechanisms and problems associated with it. Advances in biotechnology opened a door for overcoming some of the biological barriers associated with the introgression. There is still a need of developing more techniques to successfully transfer diversity without adversely affecting the economically important traits. Van Esbroeck and Bowman (1998) indicated that cotton germplasm resources would remain underutilized if new and improved methods were not developed to transfer useful allelic variation to primary cotton gene pools. It would be also helpful if breeders understand the value, availability and importance of these indispensable genetic resources.

A study was conducted to understand the measure of genetic variation present in the primitive race accessions using SSR markers at the molecular level and to understand the degree to which the genotype of the photoperiodic parent is recovered in the process of repeated

backcrossing to plant introductions. It was found that germplasm accessions had approximately 75 percent of alleles common with the *G. hirsutum* standard TM1. It was also found out that much of variation was lost during the process of recovering the recurrent parent attributing that to events such as inadvertent selection, variable recovery of alleles, heterogeneity of the collections and linkage drag (Liu et al., 2000). Campbell et al. (2009) reported that the combination of alternative breeding methods like random intermating, modified backcrossing and composite crossing along with having the novel germplasm foundation in the breeding program helped in maintaining the genetic diversity in each program.

Incorporating genetically diverse parents in breeding may help in improving the genetic base of cultivars and could possibly solve the problem of genetic vulnerability (Rodgers et al., 1983). Incorporating genetic variability may increase the chances of producing transgressive segregates for yield (Cowen and Frey, 1987). Using genetic material from different phylogenetic clusters as a parent material could improve the genetic variability. Checking the pedigree of the cultivars before using them as parent material in cultivar development may avoid the repeated use of the same breeding material (May et al., 1995). Utilizing exotic germplasm, which has not been subjected to selection, could be a better approach to enrich and improve genetic variability (Meredith, Jr., 1991).

Exotic germplasm and its utility in cotton:

Exotic germplasm usually refers to wild and feral relatives, plant introductions, landraces (from other agro-climatic zones), obsolete varieties (from other agro-climatic zones), and any other unadapted germplasm. Reports indicate that there is plenty of genetic diversity available in the form of wild and feral relatives in each of the five tetraploid *Gossypium* species (Bowman et al., 1996, 1997; Calhoun et al., 1997). The major limiting factor that limits the usage of wild and

primitive accessions in cotton breeding are photoperiod sensitivity and lack of adaptability of these accessions (McCarty et al., 1996; Van Esbroeck and Bowman, 1998). Due to photoperiod sensitivity these accessions do not flower in the long day conditions of the intended area of adaptation (Abdurakhmonov et al., 2007). Introgressing day neutral genes into plant introductions can increase the availability of these genetic resources. Blocks of genes which tend to stay together during backcrossing also makes it extremely difficult to transfer germplasm from these primitive accessions. Successful introgression of day neutral genes facilitated the cultivation of upland cotton cultivars in new areas where it would not have been an option before (McCarty et al., 1979; Zhong et al., 2002).

Primitive germplasm accessions can be considered a source of novel QTL alleles (Tanskley and McCough, 1997). Several studies have indicated the importance of primitive race accessions as a rich source of genetic variability and its utility in improving the genetic base of upland cotton (Basal et al., 2003, 2005; McCarty et al., 1998a, 1998b, 2004a, 2004b; Meredith, Jr., 1990). Many desirable breeding traits are available in converted day neutral primitive race accessions that were collected from different geographical regions. These can be useful in enriching the cotton germplasm pool (McCarty et al., 2004a, 2004b, 2007).

McCarty et al. (1979) and McCarty and Jenkins (1993) have converted 97 primitive accessions into day neutral accessions, which served as a rich breeding material for successive research studies. McCarty et al. (2007) reported that these derived primitive race accessions contain genes with significant additive genetic effects for fiber properties and also non-significant additive effects for yield improvement. Contrary to what many people think, McCarty et al (1996) indicated that exotic introgression does not necessarily incur serious losses on agronomic performance of the cultivars. Some reports indicate that introgressed primitive

accessions served as genetic reservoirs for various genes conferring resistance to pests like boll weevil (*Anthonomous grandis* Boheman), *Fusarium* wilt (*Fusarium oxysporum* f. sp. *vasinfectum* (Atk.) Snyd & Hans) and tarnished plant bug (*Lygus lineolaris* (P. deB.) (Jenkins, 1979; McCarty et al., 1986). According to Tanksley and McCough (1997) there could be novel QTL alleles available in wild primitive accessions which can significantly improve yield, if introgressed carefully.

Land races of upland cotton and wild tetraploid species were successfully utilized in transferring morphological traits like nectariless trait and glabrous leaf traits (Meyer and Meyer, 1961; Percy and Kohel 1999). Male sterile cytoplasm was introduced to *G. hirsutum* from *G. harknessii* (Meyer, 1974). Many genes conferring resistance to pests and diseases were also transferred successfully using landraces. Some major genes transferred into upland cotton were resistance to bacterial blight (*Xanthomonas campestries* pv *malvacearum* (Smith) Dye), boll worm (*Heliothis zea* (Boddie)) resistance and *Fusarium* wilt resistance (Meredith, Jr., 1991; Stewart 1994). A trait for natural defoliation in cotton was introgressed from exotic germplasm (Abdurakhmonov et al., 2005). Keeping exotic introgression breeding in mainstream breeding programs and maintaining genetic variability can provide buffering potential for the cotton genetic pool. It may translate into high reward and ready to use genetic material when an alternative source is needed (Paterson et al., 1991; Zeng et al., 2007).

With increasing demand for high quality fibers, exotic germplasm introgression can be looked as a potential source to bring in the variability for yield and fiber traits (Zeng et al., 2007, 2011). McCarty et al. (2007) used chromosomal substitution lines, converted day neutral lines, and germplasm lines derived from wild species to study the effects on agronomic and fiber properties. Results revealed significant additive effects for all traits evaluated except seed cotton

yield and fiber elongation; significant dominance effects were observed for all traits (McCarty et al., 2007; Wu et al., 2010). This study also helped to identify parents with desirable additive effects for both agronomic and fiber traits that could be utilized for multiple trait improvement.

1.1 Agronomic traits in cotton:

Days to first flower:

Days to first flower (DFF) is the number of days from planting to appearance of the first open flower in a plot. First flowering in cotton generally occurs around 56 days after planting under normal circumstances, however, it can vary depending on the cultivar, growth conditions and photoperiod sensitivity/insensitivity (Ritchie et al., 2007). Studies indicate DFF is a highly heritable trait with broad sense heritability values of 0.96 (Bhatooli et al., 2010), and 0.75 (Abbas et al., 2013). Days to first flower has been frequently associated with earliness as it is easier to work with than other earliness indicators. Days to first flower is an easily recognizable trait so chance of subjective error would be less. According to Richmond and Radwan (1962), DFF was significantly correlated with a popular earliness estimation method (ratio of weight of seed cotton harvested at the first picking to total weight of seed cotton harvested and expressed as percentage). Godoy and Palomo (1999) reported a significant correlation between DFF and percentage of crop harvested during the first picking. Cabangbang et al. (1989) concluded that DFF is a more accurate and inexpensive method of estimating earliness than any other phenological attribute of earliness.

As it was discussed earlier, photoperiod sensitivity or insensitivity could play a major role in influencing days to flowering within unadapted germplasm. The majority of exotic germplasm accessions are considered to be photoperiod sensitive which makes it difficult to flower in the production environment of U.S. cotton cultivation (McCarty et al., 2004a; 2004b).

Photoperiod sensitivity associated with flowering habit can be stated as one of the main reasons why exotic germplasm has been underutilized in cotton breeding programs (Abdurakhmonov et al., 2007). To overcome this barrier, significant effort has been invested into converting primitive accessions into day neutral accessions. Successful introgression of day neutral genes into primitive accessions has been reported (McCarty and Jenkins, 1993; McCarty et al., 1979). Since our study involves exotic lines measuring days to first flowering can provide meaningful information necessary to predict earliness in our experimental material.

Bolls plant^{-1}:

Among all the yield components, boll number is one of the major contributors to lint yield (Bridge et al., 1971; Moser, 1999; Ramey, Jr., 1972; Zeng and Meredith, Jr., 2009b). Boll number was reported as a top contributor for yield followed by seeds boll^{-1} and lint seed^{-1} (Maner et al., 1971; Worley et al., 1974). Earlier in cotton production when manual picking was the main method of harvest greater boll size was preferred over higher boll number due to the time and effort involved in hand picking. Since the advancement of mechanical harvesting increased number of bolls with higher lint percentages has become an important breeding objective. In a study comparing obsolete cultivars to modern cultivars, significantly higher yields recorded in modern cultivars was attributed to increased boll number (Ramey, Jr., 1972).

Reports indicate that boll number heritability estimates are quite variable. Lu and Myers (2011) reported that boll number per plant had the lowest heritability value among all the traits they studied in the diallel analysis using 10 most influential upland cotton cultivars in Australia. They reported a narrow sense heritability value of 0.15 and a broad sense heritability value of 0.21. Desalegn et al. (2009) reported a broad sense heritability value of 0.59 for boll number in diallel crosses they performed and considered this trait as moderately heritable. In a correlation

study conducted by Abbas et al. (2013) a broad sense heritability value of 0.77 was reported for boll number. These values may suggest that success of selection for this trait can range from ineffective to very effective and is probably population-dependent. Another important aspect of this trait is its relationship to other yield traits. Yield components are generally interdependent; changing one trait may have a positive or negative effect on other yield component traits. Results from earlier studies indicate that boll number is positively correlated with lint percentage (0.72), and negatively correlated with boll size (-0.88) and seed index (-0.91) (Lu and Myers, 2011). Worley et al. (1974) reported boll number was negatively correlated with seeds per boll (-0.12), and lint per seed (-0.08), but it was very highly correlated with lint yield (0.96). Worley et al. (1976) reported boll number accounted for 85% (R^2) of variation for lint yield. Zeng and Meredith, Jr., (2009b) observed that the average boll number in John Cotton (JC) exotic introgression lines and cultivar checks was not significantly different.

Boll size:

Boll size is another major yield component with a significant impact on yield. Boll size is a function of seed weight and lint weight, of which seed weight is the major portion of boll size. Earlier in the cotton history boll size was considered an economic trait because of less labor requirement to pick a bale from bigger bolls compared to smaller bolls. Advances in mechanized harvest machinery caused breeders to put less emphasis on this trait. In a study comparing performance of obsolete cultivars vs. modern cultivars, it was reported that boll size of obsolete cultivars was significantly larger than modern cultivars at that time (Bridge et al., 1971). A study by Culp and Harrell (1975) similarly suggested that modern cultivars have smaller boll sizes. Zeng and Meredith, Jr., (2009b) observed that the average boll size in John Cotton (JC) introgressed lines and cultivar checks was not significantly different.

Selection for boll size can be effective because boll size was reported as a moderate to highly heritable trait (Lu and Myers, 2011; Percy et al., 2006). However, it is important to point out that heritability values reported by some earlier scientists were quite variable. Some of the heritability values reported earlier were 0.22 (h^2 narrow sense (ns)) by McCarty et al. (2007), 0.22 (h^2ns) and 0.57 (H^2 bs) by McCarty et al. (2004a, 2004b), 0.40 (h^2ns) and 0.57 (H^2bs) by Bhateria et al. (2006), 0.62(h^2ns) by Desalegn et al. (2009) and 0.87(H^2bs) by Percy et al. (2006) etc. As it was mentioned earlier, boll size is reported to be negatively correlated with boll number, which makes breeding for simultaneous improvement in these two traits quite challenging. Identifying a combination with increased boll number and increased boll size would have the potential improve lint yield significantly. Earlier reports have also indicated bigger boll cultivars generally had a tendency to have larger seed in obsolete cultivars (Bednarz et al., 2006; Bridge et al., 1971). Some earlier studies indicated that boll size along with boll number were major contributors to heterosis for yield (Meredith, Jr., and Bridge, 1972; Tang et al., 1993a). Many studies have reported correlations of boll size with other yield related and fiber property traits. Lu and Myers (2011) observed a highly significant positive correlation between boll size and seed index (0.90), and a significant negative correlation with lint percent (-0.73). Boll size was reported as a medium to highly heritable trait.

Seeds boll^{-1}:

Seed boll^{-1} is another important yield component that affects seed cotton yield and lint yield. Number of seed boll^{-1} is dependent on boll and seed size. Culp and Harrell (1975) reported that selecting for medium size bolls with higher seed number while maintaining higher lint percentages can lead to increase in lint yields. Lint is an epidermal outgrowth of seed, so increasing seed number may increase the surface area available for lint development leading to

increase in lint yield (Harrell and Culp, 1976). Since seed cotton yield is a function of seed yield and lint percent, it is logical that seed boll^{-1} must be very important in determining seed cotton yield. Bridge et al. (1971) reported modern cultivars have more seed boll^{-1} compared to obsolete cultivars. Zeng and Meredith, Jr., (2009b) observed that the average of number seeds per boll in JC introgressed lines were not significantly different from cultivar checks.

During the course of cotton domestication and improvement, intentional selection for higher seed number likely may not have been an objective, but may have been increased as a byproduct of selection for other traits. Selecting for higher seed number may have negative impact on other traits. Seeds per boll is negatively correlated with lint per seed (0.49), which indicates an increase in seeds per boll may reduce lint per seed (Worley et al., 1974). Smith and Coyle (1997) reported seed number and fiber strength to be negatively correlated, and increasing one trait would cause a decline in the other. Harrell and Culp (1976) indicated that the negative correlation between fiber strength and seeds per boll might not be absolute, because several instances have occurred in the past where both high fiber strength and more seed number per boll were improved simultaneously. Selection for number of seed boll^{-1} may also improve other traits. Desalegn et al. (2009) reported significantly high positive correlations of seeds per boll with seed cotton yield, bolls per plant, and lint yield. They also reported a medium correlation with lint percentage, and low correlations with lint index, seed index and boll size.

Lint seed^{-1}:

Lint is an epidermal single celled outgrowth of the seed coat. Lint seed^{-1} or Lint Index (when expressed as mass for 100 seeds) is a good indicator of the amount of lint fiber on each seed. Worley et al. (1976) indicated that the most basic component of lint yield must be lint mass per seed. This trait is determined by the number of fibers per seed, length of the fiber and

unit length of the fiber (Worley et al., 1974). Since lint mass per seed is a function of lint fibers and seed size and number, factors affecting any of these traits may positively or negatively influence this trait. As mentioned earlier, increasing seed surface may offer more area for lint fiber development. Harrel and Culp (1976) inferred that lower lint mass per seed might be the reason behind lower lint percentages in most of the Pee Dee lines used in their study.

Like most other yield related traits, breeding for lint mass per seed may positively or negatively influence other traits. Earlier reports have indicated low correlation values between lint per seed and lint yield exist in cotton (Worley et al., 1974). The correlation between lint per seed and lint yield may have been increased during the course of cotton improvement. Correlation values reported for lint index with other yield traits were significantly higher for seed cotton yield, boll weight, lint yield, lint percentage, and seed index, whereas non-significant negative correlation values were reported for lint index with boll weight and seeds per boll (Desalegn et al., 2009; Moser, 1999; Smith and Coyle, 1997). Since improving this trait may have a positive impact on other economic traits, mainly lint yield, efforts to improve lint mass per seed should be pursued. Exotic germplasm may offer some options for improving this trait, and may also help in breaking linkages with negatively correlated traits. Earlier reports indicating success in identifying lines with improved lint mass per seed using exotic germplasm offers hope for improvement. Zeng et al. (2010) reported in their study that two out of four exotic germplasm lines derived from John cotton germplasm outperformed two cultivar checks with respect to lint mass per seed. But in another study, mean lint mass per seed of the selected exotic germplasm lines (species polycross) was not significantly different from the mean of five cultivars checks (Zeng and Meredith, Jr., 2009a).

Seed cotton yield:

Seed cotton yield is a complex trait determined by a number of individual components. It can be affected by traits such as boll number, boll size, seeds per boll, and lint per seed (Wells and Meredith, Jr., 1984; Zeng and Meredith, Jr., 2009a). Since lint yield is a product of seed cotton yield multiplied by lint percentage, improving seed cotton yield may also lead to increases in lint yield provided lint percentages are maintained. Seed cotton yield in general is perceived as a trait highly influenced by environmental conditions. Reported heritability values bear this out as seed cotton yield has a low heritability (Desalegn et al., 2009; Jenkins et al., 2009).

Seed cotton is determined by many yield components and associated with many other traits, so knowing these associations and modifying these components may lead to a higher success rate in yield improvement. Seed cotton yield was reported to have a significant positive correlation with bolls per plant, boll weight, lint yield, lint percent, lint index, and seed per boll (Desalegn et al., 2009; Khan et al., 2010). One of the major setbacks in improvement has been the negative linkages between yield and fiber properties. Improving seed cotton yield while simultaneously maintaining fiber properties has been quite a challenge because of these negative associations. However, some studies have found a significant positive correlation between seed cotton yield and fiber strength, and seed cotton yield and fiber fineness (Azhar et al., 2004). Exploring genetic diversity within *G. hirsutum* seems like an attractive option to break the negative linkages with seed yield. Some researchers have had success in identifying lines with good fiber properties and acceptable yield using exotic germplasm. Wu et al. (2010) have identified some lines developed using exotic germplasm that performed better than cultivar checks used as parents.

Lint percentage:

Lint percentage is the proportion of lint weight in the cleaned seed cotton expressed as percentage. Lint percentage is an important trait because of its direct impact on lint yield. Meredith, Jr., (1984) reported that lint percentage contributed 70 to 90 % of variation for lint yield. Lint percentage was reported as a critical factor in maintaining higher yield as it is determined by boll size, seeds per boll and lint mass per seed (Culp and Harrell, 1975). During the course of cotton domestication and improvement, significant improvement in lint yield has been attributed to improvements in lint percentage (Bridge et al., 1971). An increase in lint percentage along with an increase in boll number almost guarantees higher yields; this has been observed in some modern day cultivars (Bridge and Meredith, Jr., 1983; Bridge et al., 1971; Culp and Green, 1992). Lint percentage was reported to be a highly heritable trait and stable across environments (Desalegn et al., 2009; Lu and Myers, 2011; Percy et al., 2006; Tang et al., 1993a; Ulloa, 2006;). Selection for this trait can be effective because of its high heritability (Meredith, Jr., and Bridge, 1973). Miller and Rawlings (1967) reported a significant improvement for lint percentage with each cycle of recurrent selection for that trait.

Like other yield components, lint percentage also has positive and or negative interactions with other traits. Lint percentage and lint yield were reported to have a highly significant positive correlation (Desalegn et al., 2009; Culp and Harrell, 1975). Lint percentage is positively correlated with seed cotton yield, bolls per plant, lint index, and seeds per boll and negatively correlated with seed index (Desalgen et al., 2009; Lu and Myers, 2011; Stewart and Kerr, 1974; Zeng et al., 2007). The main challenge to overcome in the improvement of lint percentage is the negative association with some of the economically important fiber properties. Wang et al. (2011) reported a strong negative correlation between lint percentage and fiber length and fiber strength. Similar correlations were also reported by Lu and Myers (2011),

however the only significant negative correlation they found was with fiber strength and the correlation with fiber length was not significant. Exploring the use of exotic germplasm may offer some possibility in breaking these unfavorable negative linkages between lint percent and fiber properties. Introgressing exotic genes into *G. hirsutum* L. strains followed by 11 generations of random mating has helped in isolating improved SP lines (species polycross lines) with better lint percentage than the cultivar checks used in an earlier study (Zeng and Meredith., Jr., 2009a).

Lint yield:

Lint yield is the most important economic trait in determining the value of a cotton crop and cultivar worth. During the course of cotton history significant improvement has been made in lint yield. The success in improvement can be attributed both to breeding efforts (genetics) and improvements in crop management and harvest technology. Since lint yield is a product of seed cotton yield and lint percentage, simultaneous improvement in these two traits might lead to increased yield. Lint yield has long been known to be a complex trait that is determined by number of individual traits such as boll number, boll mass, seeds per boll, lint per seed and lint percentage (Coyle and Smith, 1997; Wells and Meredith, Jr., 1984; Worley et al., 1976; Zeng and Meredith, Jr., 2009a, 2009b). Earlier reports have indicated that boll number and lint percent are the most significant contributors to the lint yield among all yield components (Meredith, Jr., 1984; Worley et al., 1976). Significant influence of these two traits on lint yield has also been found be other researchers (Bridge et al., 1971; Culp and Green, 1992; McCarty et al., 2006; Ramey, Jr., and Worley, 1973). Therefore it is critical to understand relationships with other traits to efficiently improve lint yield.

Lint yield has been reported as a low to moderately heritable trait. Some of the heritability values reported in earlier studies were 0.45 (Murray and Verhalen, 1969), 0.48 (Meredith, Jr., and Bridge, 1973), and 0.72 (Desalegn et al., 2009). Selection for lint yield may also lead to improvement in other yield related traits. Miller and Rawlings (1967) reported that selection for lint yield has improved lint percentage, seeds per boll, earliness, fiber elongation, and decreased fiber fineness with each cycle of recurrent selection. Lint yield was reported to have highly significant positive correlation with seed cotton yield, bolls per plant, lint percent, lint index, seed per boll, and non significant correlations with boll weight, and a negative correlation with seed index (Desalegn et al., 2009; Percy et al., 2006). One of the major challenges that breeders have to overcome is the negative linkage between yield and fiber quality (Miller and Rawling, 1967; Zeng et al., 2007). Exploring genetic diversity available in the cotton gene pool may be the only viable option in identifying and selecting lines with both improved yield and improved fiber properties. Expanding the genetic base could provide a solution by introducing new genes into the adapted germplasm pool and increasing chances of new gene combinations which may be better in terms of both yield and fiber properties (Zeng and Meredith, Jr., 2009a, 2009b). Using plant introductions to introduce new genetic variability may be a suitable option, as some earlier studies report success in this area. John cotton populations, which were developed by introgressing exotic genes, have helped in deriving lines, which are good in fiber properties with acceptable yields (Zeng, et al., 2010). Similar kinds of success were also reported in species polycross populations, which were also developed by introgressing exotic genes (Zeng and Meredith, Jr., 2009a).

Earliness:

Earliness can be defined as the shortest amount of time to produce a profitable crop (Guthrie et al., 1995). The genus *Gossypium* is characterized by an indeterminate flowering and fruiting habit. The practical implications of indeterminate flowering/fruiting type may not fit well in a modern production system. Producers prefer cultivars that mature predictably in a set amount of time to facilitate their operations being performed on a specific timetable. Earliness is an important trait to study, because bolls developing at different times are reported to have different fiber properties (Meredith, Jr., and Bridge, 1973). Early maturing cultivars have a shorter life cycle, which helps plants to avoid unfavorable situations that might occur during the growing season. It offers advantages such as maximizing the utilization of early season moisture, escaping pests, increased chances of recovering from stress, and escaping late seasons stress conditions like frost (Braden and Smith, 2004; Murray and Verhalen, 1969).

Earliness can be estimated in many different ways, but the most popular method is by calculating the ratio of weight of seed cotton yield harvested in the first picking to the total weight of seed cotton harvested at the end of the season and expressed as percentage (Bourland et al., 2001; Richmond and Ray, 1966). Earliness was reported as a moderate to highly heritable trait with a heritability value of 0.73 (Murray and Verhalen, 1969). Selection for earliness can be effective, and response to selection has showed a significant improvement with each cycle of recurrent selection (Miller and Rawling, 1967). Culp and Green (1992) indicated that selection for early maturing genotypes was one of the main reasons for increases in yield potential in modern cultivars. One of the main setbacks could be its negative correlation with fiber quality. Since early maturing varieties takes less time to mature, the amount of time available for boll maturation is also less, which may result in poor fiber properties (Braden and Smith, 2004).

Lodging:

Lodging resistance is a desirable trait in any cotton breeding program, but it is especially important where wild relatives, exotic germplasm or other wide sources are used. Lodging resistance is an important trait where fields are frequently exposed to high winds. In the United States, lodging resistance can be considered as one of the important traits due to mechanized harvesting (Acquaah, 2007). Cotton is naturally a perennial plant with an indeterminate growth habit, and generally grows to about 1.5 meters in height. However cotton is cultivated as an annual and tall plant height is not a preferred trait. The strength of the plant stem should be sufficient to bear the weight of bolls so lodging resistance is an important trait. Modern cultivars are quite resistant to lodging and have the ability to support more bolls per unit production area (Schwartz and Smith, 2008). Plant introductions generally have a tendency to grow taller when introduced to new climatic conditions to which they are not adapted. Studies in soybean, where exotic germplasm has been used as a source of introgression, recorded increased lodging percentages with increases in exotic germplasm (Schoener and Fehr, 1979; Vello et al., 1984). However, since cotton stem stature is different from soybean, the severity of lodging may not be the same.

1.2 Fiber properties in cotton:

Fiber length:

Fiber length is one of the most important fiber quality traits and affects processing and yarn quality. It is the average length of the longest 50 percent (upper half mean length) of the cotton fibers. Fiber length is important for textile processing because longer fibers produce stronger yarns by allowing fibers to coil around each other for more times. Fiber length for a major part is controlled by genetic component, but it is also influenced by environmental conditions like extreme temperatures, water stress, nutrient deficiencies and also mechanical

conditions like extreme cleaning or drying (Bradow and Davidonis, 2000; Cotton Incorporated, 2011). Genetic improvement is an effective approach for improving fiber length (Campbell et al., 2013; Zeng and Meredith, Jr., 2009b). While fiber length is largely governed by genetic component, genotype × environment also plays a significant role in expression of this trait up to some extent (Zeng and Meredith, Jr., 2009a; Zeng et al., 2010, 2011).

Fiber length is associated with other fiber properties such as fiber uniformity, fiber strength, fiber uniformity and fiber elongation. Reports indicate fiber length and fiber strength are positively correlated, implying improving fiber length also improves fiber strength (Lu and Myers, 2011; Ulloa, 2006; Zeng and Meredith, Jr., 2009b). One of the major challenges with improving fiber length is its negative correlation with lint yield (Bridge et al., 1971; Campbell et al., 2012; Meredith, Jr., and Bridge, 1971). However, the strength of negative correlation between fiber length and lint yield has been reduced through breeding efforts in Pee Dee germplasm (Campbell et al., 2011, 2012, 2013). Fiber length is also highly correlated with lint percentage (Smith and Coyle, 1997; Zeng et al., 2007). Expanding genetic diversity may help in enhancing favorable combinations and eliminating negative linkage blocks. Breeding efforts using exotic germplasm for introgression have resulted in the development of John cotton and species polycross lines, where successful lines with improved fiber strength were achieved (Zeng and Meredith, Jr., 2009a, 2009b; Zeng et al., 2010).

Fiber strength:

The importance of fiber strength increased profoundly with technological advancements in spinning technology. Modern spinning machinery requires higher fiber strength for improved efficiency and product quality. Fiber strength is the amount of force required to break a one tex unit in size (a tex unit is equal to the weight in grams of 1000 meters of fiber). The most

efficient way of improving fiber strength is through genetics and breeding; only a few genes control fiber strength (Meredith, Jr., 2003, 2005). While fiber strength is largely governed by genetic component, genotype × environment also plays a significant role in the expression of this trait (Cotton Incorporated, 2011). Reports from exotic introgression studies provide evidence for this hypothesis (Zeng and Meredith, Jr., 2009a, 2009b; Zeng et al., 2007, 2011). One of the problems associated with improvement of fiber strength has been its negative association with some yield components, mainly lint yield. A negative association of fiber strength with lint yield has been reported in several studies (Bridge et al., 1971; Campbell et al., 2012, 2013; Culp et al., 1979; Green and Culp, 1990; McCarty et al., 1996; Meredith, Jr., 2005; Meredith, Jr., and Bridge, 1971; Zeng and Meredith, Jr., 2009b). Expanding genetic variability may help in breaking these negative linkage blocks (Culp et al., 1979; Miller and Rawlings, 1967;). Successful breeding with increased genetic gains in agronomic traits while maintaining fiber strength has been reported in Pee Dee germplasm breeding program (Campbell et al., 2011, 2012, 2013; Culp and Harrell, 1977; Culp et al., 1979). Reports also indicate that fiber strength is highly correlated with lint percentage, boll weight, seed weight and 2.5% span length (Meredith, Jr., 2005; Miller and Lee, 1964; Worley et al., 1976). In John cotton germplasm lines derived using exotic germplasm significantly higher fiber strength was observed compared to cultivar checks (Zeng et al., 2010).

Micronaire:

Fiber fineness is an important fiber property that plays an important role in spinning performance. Fiber fineness is related to the texture of fiber and denotes the size of the cross section dimensions of the fiber. If the same count of yarn is spun from two samples, the sample with finer fibers will have more number of fibers in cross section which makes it stronger than

the sample with less fine fibers (Cottonguide, 2015). Cotton fibers that are strong and fine are considered better for processing and help in better yarn distribution. Coarser cotton fibers are generally undesirable for spinning industry as they affect processing speeds. Fiber fineness is commonly measured using the air-flow method and is expressed in micronaire units. Low micronaire value indicates fine cotton fibers whereas high micronaire value indicates coarser cotton fibers. Micronaire values which ranges between 3.5 and 4.9 are considered as fine cottons; micronaire values ranging between 4.3 through 4.9 receive a premium (Cotton Org, 2015). Fiber fineness is influenced by both genetic and environmental factors (Cotton guide, 2015). However, environment plays a substantial role therefore parental performance is a weak predictor of fiber fineness (Campbell et al., 2013). Meredith, Jr., (2003) reported that the environmental component accounted for 61 percent of total variation in an analysis of 36 years of high quality cultivar testing data. Similar results were also observed in Pee Dee germplasm collection following seventy years of plant breeding, environmental component ranged up to 82 percent for micronaire (Campbell et al., 2012). Several studies involving exotic germplasm introgression have indicated significant genotype, environment, and genotype × environment interactions for fiber fineness (Zeng and Meredith, Jr., 2009b; Zeng et al., 2007, 2010, 2011).

Fiber fineness is also associated with agronomic and fiber properties. Bridge et al. (1971) reported that the lowest yielding cultivars also had the lowest micronaire values. Ulloa (2006) reported micronaire is positively correlated with lint percentage and boll weight and negatively correlated with fiber elongation. Micronaire values were reported to be positively associated with lint weight within a boll (Smith and Coyle, 1997). Lu and Myers (2011) reported fiber fineness is significantly correlated with every yield component except lint index. Breeding efforts using exotic germplasm for introgression have resulted in the development of John cotton

lines, where successful lines with improved fiber fineness were achieved (Zeng and Meredith, Jr., 2009b; Zeng et al., 2010).

Fiber uniformity:

Fiber uniformity is one of the three important fiber properties besides micronaire and fiber strength that determine the quality premium paid for cotton (Cotton Org, 2015). In HVI analysis fiber uniformity is the ratio between the mean length and upper half mean length of the fibers and expressed in percentage, whereas in fibrograph estimation, it is the ratio between 2.5% span length and 50% span length (Bradow and Davidonis, 2000; Cotton Incorporated, 2011). If all the fibers in a sample are of equal length, the mean length and upper half mean length would be equal and fiber uniformity index would be 100 percent. Uniformity in fiber length is an important trait that affects yarn and fabric processing. Fibers with poor uniformity can significantly impact spinning process in a negative way as the low fiber uniformity index values indicate higher short fiber content (Cotton Incorporated, 2011). Fiber uniformity value is a good predictor of short fiber content, because low uniformity value likely indicates high short fiber content. Yarn strength, evenness and efficiency of the spinning process are influenced by uniformity index. According to Lu and Myers (2011) fiber uniformity is positively correlated with fiber length and fiber strength.

Fiber elongation:

Fiber elongation is a measurement of elasticity (stretch before breaking) expressed in percentage. Fiber elongation enables all the fibers to share in contributing to yarn strength. Benzina et al. (2007) reported that the combination of increased fiber tenacity and decreased elongation resulted in less energy required to break yarn. The same authors also reported that an increase in elongation resulted in significant increase in work required to break yarn. Faulkner et

al. (2012) reported a strong positive correlation (r = 0.84) between yarn elongation and yarn work to break. Fiber elongation is determined by cultivars and environment (Bridge et al., 1971; Ng et al., 2014; Zeng and Meredith, Jr., 2009b; Zeng et al., 2007, 2010, 2011). Exotic germplasm introgression has resulted in improved germplasm lines with significantly higher elongation percentages than cultivar checks in John cotton germplasm lines (Zeng and Meredith, Jr., 2009b; Zeng et al., 2010).

Short fiber content:

Short fiber content is an important fiber property resulting in fiber waste. It also leads to weaker yarn (Ulloa, 2006). Short fibers have been defined as those measuring less than 12.7 mm in length (Bradow and Davidonis, 2000). Shorter fiber is not a desirable trait as it creates processing problems as well as quality issues and reduces spinning efficiency. If the content of short fiber is high, more fiber is required in making yarn. Excessive cleaning and drying in order to achieve an industrial bale grade leads to more short fiber content (Cotton Incorporated, 2011). An increase in short fiber causes spinning end breaks, processing waste, floating fibers, formation of neps, and yarn unevenness. According to May and Jividen (1999) breeding progress in alleviating short fiber content is limited due to its low heritability; however, significantly high genotype effects for short fiber content was observed in other studies indicating breeding progress may not necessarily be slow (Zeng and Meredith, Jr., 2009a; Zeng et al., 2007). Exotic introgression studies have reported significant genotype and environmental effects for short fiber content (Zeng et al., 2010, 2011; Zeng and Meredith, Jr., 2009b). Exotic germplasm introgression has resulted in improved germplasm lines with significantly lower fiber content than cultivar checks (Zeng et al., 2010).

GENERAL OBJECTIVES

1. To study the effect of exotic germplasm on the agronomic traits of local adapted cultivars
2. To study the effect of exotic germplasm on the fiber properties of local adapted cultivars

2. Effect of Exotic Germplasm on Agronomic Traits

ABSTRACT

Upland cotton (*Gossypium hirsutum* L.) is an economically important natural fiber crop in the world. Along with fiber, it is also valued for its oil and protein portion of the seed. Upland cotton is reported to be facing a risk associated with a narrow genetic base. The utilization of closely related parents and many reselections within elite cultivars in the development of cultivars has led to a narrow genetic base. Genetic uniformity predisposes crops to various abiotic and biotic stresses. Cotton cultivars with increased genetic diversity can offer plasticity to be able to respond to stressful environments. Exploration of exotic germplasm for introgressing novel genes and broadening the genetic base of cotton may offer additional genetic variation to help future trait improvement. Our research is focused on the objective of determining the effect of exotic germplasm on agronomic traits of adapted cotton cultivars. Two plant introductions (TX245 and TX1419) were systematically crossed with four cultivars (FM966, PM1218, Deltapearl and SG747) to derive five groups of lines with 0, 25, 50, 75 and 100 percent exotic germplasm. Experimental materials were tested in a randomized complete block design (RCBD) and analyzed using PROC MIXED as implemented in statistical analysis software (SAS 9.2).

Results revealed highly significant effects due to population, exotic percentage, and population × exotic percentage interactions for all the agronomic traits except for boll plant^{-1}, where the effect due to exotic percentage was not significant. Genotype × environment interactions were also significant for most of the traits. An increase in exotic parentage percentage significantly lowered agronomic and fiber properties, however the magnitude of

effect varied with populations and exotic parentage percentage levels. Among agronomic traits, seed cotton yield, lint seed^{-1}, lint percentage and lint yields were the most affected, whereas days to first flower, bolls plant^{-1} and lodging were traits least affected with an increase in exotic percentage. Boll size and seeds boll^{-1} were unaffected or improved in populations (except in POP1) derived from exotic parent TX245, whereas both traits declined significantly in populations derived from exotic parent TX1419. In most populations an increase in exotic parentage percentage did not show any significant difference up to 25 percent exotic germplasm introgression for all the agronomic traits except for lint mass seed^{-1}. In almost all the cases where exotic parent was significantly better, no significant difference was observed up to 25 percent suggesting an increase in exotic germplasm can expand genetic base without adversely affecting agronomic traits.

2.3 Materials and methods

This research was conducted in summer 2009 and 2010 at two locations, Tallassee, Alabama and Florence, South Carolina. The first site was located at the Plant Breeding Unit, E.V. Smith Research Center, Tallassee, AL (32°29′N, 85°53′W) and the second site at Coastal Plains Soil, Water and Plant Research Center, Florence, SC (34°29′N, 79°48′W). Soils were Wickham fine sandy loam soil (fine-loamy, mixed, semiactive, thermic Typic Hapludults) at E.V. Smith Research Center, and Goldsboro loamy sand (fine-loamy, siliceous, thermic Aquic Paleudults) at Pee Dee Research Center. Monthly averages of temperatures and rainfall for both years and locations are graphically represented in Fig 1 and Fig 2 respectively.

The exotic introgression percentage groups of lines used in this study were developed from two plant introductions and four adapted cultivars. Plant introductions TX245 (PI 165358) and TX1419 (PI 530110) were selected as exotic parents in this study, because they showed moderate levels of resistance to reniform nematode (*Rotylenchulus reniformis* Linford & Oliveria) in an earlier study (USDA, 2006; Weaver et al., 2007). TX245 is an introduction from the state of Guerrero in Mexico and belongs to the race *latifolium*. TX1419 is an introduction from the state of Bahia in Brazil; its race is not yet classified. Four cultivars FM966, PM1218, Deltapearl, and SG747 were used as adapted parents in developing advanced breeding lines. These four cultivars were selected based on their performance and adaptability and represent the elite cotton germplasm for the mid-south and southeastern USA cotton production regions.

Two plant introductions (TX245, TX1419) were systematically crossed with four cultivars (FM966, PM1218, Delta Pearl, SG747) to develop eight populations (Table 1) with five

exotic parentage percentage levels within each population. The parents and their hybrid progeny were intermated in different schemes to form five exotic percentage groups EP0, EP1, EP2, EP3, and EP4 with 0%, 25%, 50%, 75%, and 100% PI germplasm respectively. The mating scheme followed was similar to Fehr and Clark (1973). Adapted parents and exotic parents represent the 0 percent and 100 percent exotic groups respectively (Table 2). $F_{2:4}$ lines were used for 50 percent exotic, whereas 25 and 75 percent exotic were represented by $BC_1F_{2:4}$ lines. The backcross to an adapted parent represents lines for 25 percent exotic, and the backcross to an exotic parent represent lines for 75 percent exotic. Crossing work for developing these advanced breeding lines began in the year 2005 and carried to the next generation in 2006. In the year 2007, a total of 1466 lines from all eight populations (2PIs x 4 cultivars) with the above mentioned exotic percentages were planted for generation advancement at E.V. Smith Research Center, Tallassee, AL. Out of these 1466 lines, 6 lines (5 lines + 1 additional line for backup) per each exotic group per population were randomly selected and harvested. These selected lines were again planted in 2008 for seed increase and multiplication.

2.3.1 Field experiment:

Field experiments were conducted in 2009 and 2010 at two locations, Tallassee, Alabama, and Florence, South Carolina. For each of the 8 populations, 5 lines representing each exotic percent group were planted in a randomized complete block design (RCBD) with 2 years, 2 locations, 2 replications and 5 blocks as class factors. The combination of year and location was considered as environment for the purposes of statistical analysis. Plots were double-row with each row measuring 6 m long with 1 m spacing between rows. In 2009, some plots at Tallassee location were infested with nematodes and *Fusarium* spp due to the presence of inoculum left over from an earlier nematode trial conducted at the same site two years prior to

testing. The infested plots were removed from data analysis in order to reduce the noise. A total of ten agronomic traits were studied for the purposes of this experiment.

2.3.2 Data collection:

2.3.2.1 Days to first flower:

Days to first flower is the number of days from the date of planting to the appearance of first open flower. Due to logistic reasons days to first flower observations were recorded only for the Tallassee, Alabama location in both 2009 and 2010. Plots were scouted on a daily basis to record days to first flower.

Days to first flower = Date of appearance of first flower – Date of planting.

2.3.2.2 Boll plant^{-1}:

Boll plant^{-1} is the average number of bolls for each plant. A total of five different plants from each line were randomly selected and tagged. These five plants were randomly selected and tagged before flowering to avoid preferential selection. Number of bolls on all five plants were counted and divided by five to get the average number of bolls plant^{-1}. The total number of bolls harvested divided by the number of plants from which it was collected gives the average number of bolls per plant.

Boll plant^{-1} = (Total number of bolls harvested / number of plants).

2.3.2.3 Boll size:

Before machine harvest a hand-harvested sample of 50 bolls was collected from the upper, middle and lower positions of randomly chosen plants. Each 50-boll sample was weighed and recorded. Weight of 50 bolls sample divided by 50 gives the average single boll size.

Boll size (g) = (Sample boll weight/ number of bolls).

2.3.2.4 Seeds boll^{-1}:

Boll samples collected from each plot were ginned using a laboratory saw gin. Seed weights and lint weights were recorded for all ginned boll samples. Seed index (100 seeds weight in grams) was taken for each sample by randomly counting 100 seed and weighing them. By using seed index as a standard, total number of seeds in each boll sample was calculated. Total number of seeds divided by number of bolls gives seeds per boll.

Total number of seeds in a boll sample = [(Sample seed weight x100) / (seed index)].

Seeds boll^{-1} = (Total number of seeds in boll sample / Total sample boll number).

2.3.2.5 Lint seed^{-1} (g):

Lint per seed is a function of number of lint fibers per seed and the average weight per fiber (Lewis, 2001). After ginning boll samples, lint weights and seed weights were recorded for each sample. Lint weight divided by total number of seeds in the boll sample gives the average lint per seed. In our experiment it is expressed for 100 seeds.

Lint seed^{-1} (g) = (Lint weight/ total number of seeds) x 100 (for 100 seeds).

2.3.2.6 Seed cotton yield:

Each plot was spindle harvested and seed cotton yield was recorded for each plot. Plot yields were corrected by adding the 50-boll sample weight and then expressed in kg ha^{-1}. In other words seed cotton yield per plot is the sum of the 50 boll sample weight and weight of seed cotton harvested by the mechanical harvester. Seed cotton harvested from each plot is then converted into kg ha^{-1}.

SCY (kg/ha) = [((Total plot yield in kg + sample bolls weight in kg) x 10000 m^2)/ (6 m x 2m)]

2.3.2.7 Lint percentage:

Lint percentage is a calculated value derived from weight of lint after ginning. After ginning the seed cotton in each boll sample, lint weights and seed weights were recorded. Lint weight divided by seed cotton weight (lint + seed) expressed in percentage gives lint percentage.

Lint percentage = [((Lint weight) / (Lint weight + Seed weight)) x 100]

2.3.2.8 Lint yield:

Lint yield is calculated from seed cotton yield and lint percent. Seed cotton yield per acre multiplied by lint percent gives lint yield per acre.

Lint yield (kg/ha) = Seed cotton yield (kg/ha) x lint percentage/100

2.3.2.9 Lodging:

Lodging was evaluated at maturity for all plots at Tallassee, Alabama location in 2009 and 2010. Data were sorted into five different classes depending on the angle of inclination. The five different classes are L5 (all prostrate) with 0°, L4 with 22.5°, L3 with 45°, L2 with 67.5°, and L1 (all erect) with 90° angle of inclination. In other words, a lower number indicates lesser

lodging percentage and higher lodging resistance. For each plot data were recorded in such a way to represent the majority of the plants in a plot.

2.3.2.10 Earliness:

In the year 2009, earliness observations were only recorded for three populations; PG6, PG7 and PG8. Due to heavy rains we were not able to pick twice for the other 5 populations at appropriate time. Seed cotton was handpicked from each plot on two different dates. First picking was done when at least 75 percent or more number of bolls on adapted controls appeared to be ready for harvest. Second picking was done when almost all bolls appeared to be ready for harvesting. Seed cotton weight was recorded for each line from the first and second picking separately. Percent harvestable seed cotton in the first picking as a part of the total harvested seed cotton gives the estimate of earliness (Godoy and Palomo, 1999).

Earliness = [(Harvestable cotton first picking / Total harvested cotton first and second picking) × 100]

2.3.3 Statistical methods:

Data were checked for normality criteria using Statistical Analysis System SAS® PROC UNIVARIATE. Residuals were plotted in histogram and qq plots by selecting student panel option in PROC GLIMMIX module in SAS version 9.2 (SAS® institute, 2008). Mixed models analysis of variance procedure as implemented in SAS® PROC MIXED was used to analyse the data. Population, exotic percentage, and population × exotic percentage were treated as fixed effects, whereas, years, locations, and replications were specified as random effects using the RANDOM statement. The decision to choose a combined analysis over individual analysis of years and locations was chosen depending on the ANOVA results from the analysis. Mean

comparisons between exotic percentage groups and their respective controls (EP0; 100 percent exotic) within each population were performed using Dunnett's multiple comparison procedure. Tests of significance for LSMEANS was performed using slicediff option and for covariance parameters using COVTEST.

Table 1: Parents used to develop exotic percentage groups

Populations	Parents
POP1	FM966 × TX245
POP2	PM1218 ×TX245
POP3	Delta Pearl × TX245
POP4	SG747 × TX245
POP5	FM966 × TX1419
POP6	PM1218 × TX1419
POP7	Delta Pearl × TX1419
POP8	SG747 × TX1419

Table 2: Mating scheme used to develop the experimental materials

Exotic Group	Mating scheme	% Exotic germplasm
EP0	Adapted parent	0%
EP1	[(Adapted × Exotic) × Adapted] $BC_1F_{2:4}$	25%
EP2	[Adapted × Exotic] $F_{2:4}$	50%
EP3	[(Adapted × Exotic) × Exotic] $BC_1F_{2:4}$	75%
EP4	Exotic parent	100%

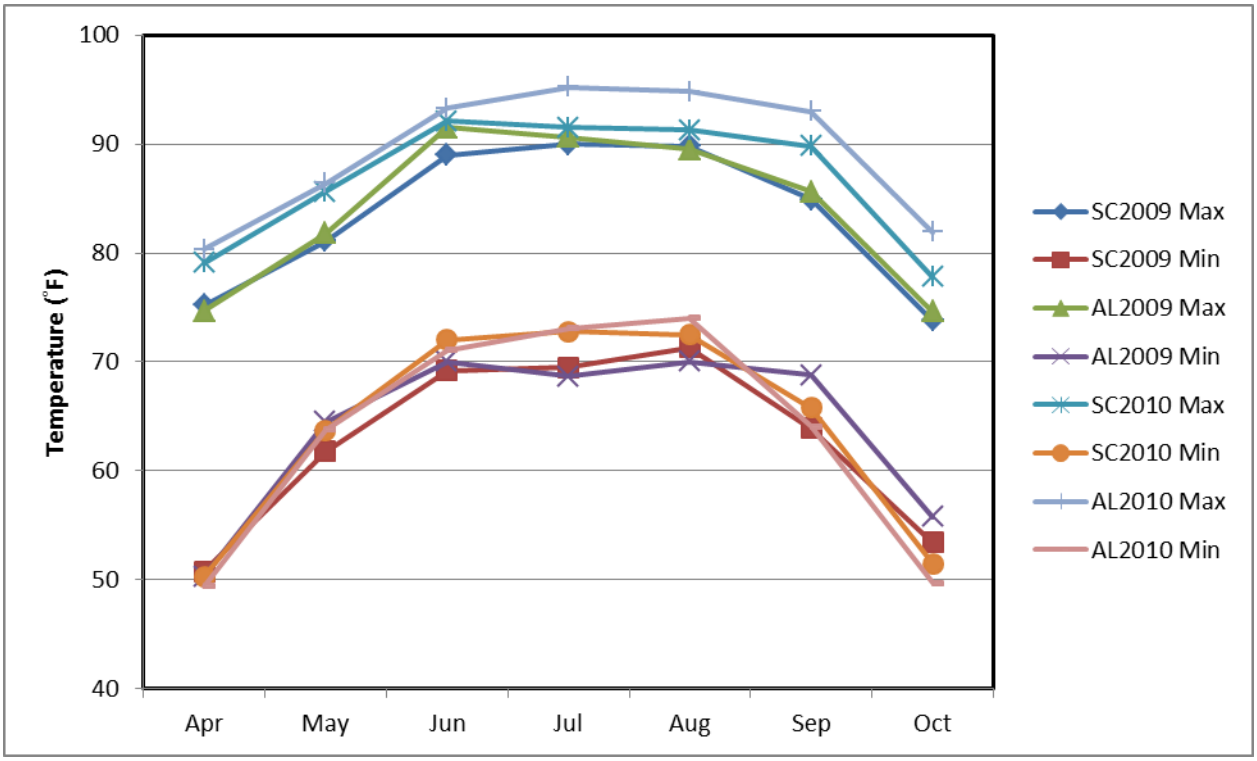

Figure 1. Monthly average temperatures (°F) during the crop seasons 2009 and 2010, at Florence, SC and Tallassee, Alabama. Source: www.awis.com (E.V. Smith climate data), www.usclimatedata.com (for Florence, SC).

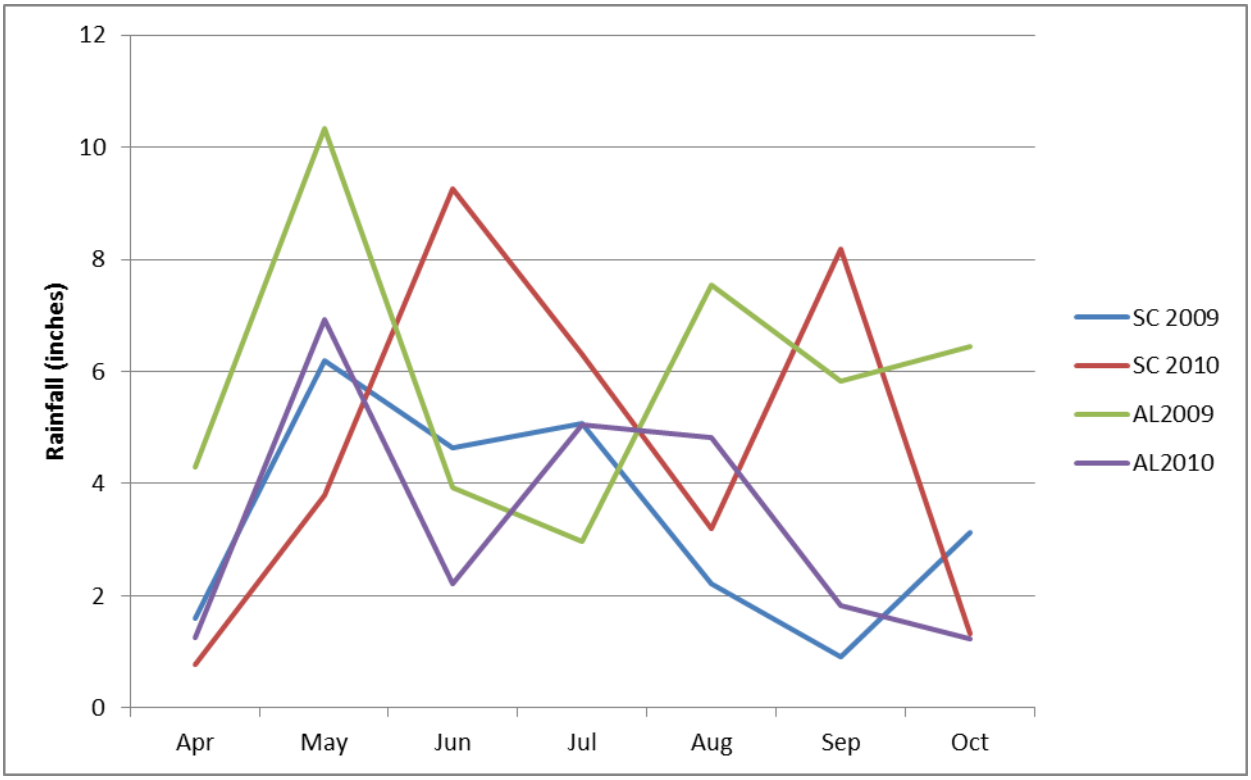

Figure 2. Monthly average rainfall during crop seasons 2009 and 2010, at Florence, SC and Tallassee, Alabama. Source: www.awis.com (E.V. Smith climate data), www.usclimatedata.com (for Florence, SC).

2.3 Results and Discussion

2.3.1 Days to first flower:

ANOVA results and covariance parameter estimates are presented in Table 3. Tests for fixed effects revealed that population had a significant effect ($P \leq 0.0387$), whereas exotic percentage, and population × exotic percentage had a highly significant effect ($P \leq 0.0037$ and $P \leq 0.0001$ respectively) on days to first flower. Covariance parameter estimates indicate (replication within year) and (exotic percentage × replication within year) were not significant. Interaction effects (population × replication within year), and (population × exotic percentage × replication within year) were highly significant. Since replication within year was not significant, interaction effect due to population may be the major source of variability in population × replication within year. In a similar way population × exotic percentage interaction must be the major source of variation in the interaction population × exotic percentage × replication within year.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 4). Except in populations POP2 and POP7, there was no evidence to show increase in exotic percentage affected days to first flower. In POP2, increase in exotic percentage beyond 50 percent significantly delayed flowering on an average of four days, whereas in POP7 increase in exotic percentage beyond 50 percent enhanced flowering on an average of three days. Graphical illustration of days to first

flowering shows a range of 52 to 57 days from the earliest to latest (Figure 3). The difference between exotic parents and adapted controls appears to be less than 3 days which indicate that the exotic parents chosen for this study were non photoperiodic, and thus the traits studied were not affected by plant photoperiod response. Since each adapted parent appears twice across eight populations, differences between the same adapted controls across experiments can be attributed to testing environment. On an average, there was a difference of up to two days observed between the same controls in different experiments. It appears that the populations derived from exotic parent TX245 showed a slightly different response from the populations derived from exotic parent TX1419. Flowering seems to be delayed with increase in exotic percentage among populations POP1-4, whereas flowering seems to be enhanced with increase in exotic percentage among populations POP5-8. However, except in POP2, those differences were not statistically significant.

From these results it can be inferred that an increase in exotic parentage percentage did not significantly influence days to first flower in these populations. The significant differences observed in POP2 and POP7 with respect to 75 and 100 percent exotic groups may have been an amplified effect of testing environment. The same adapted parent in POP2 & POP6 recorded a difference of up to 2 days, whereas the same exotic parent in POP1-4 recorded a difference of 2 to 3 days. It may be that the difference observed in 75 and 100 percent exotic is an overlapping effect of those differences. A similar kind of effect may explain the significant difference observed in 75 percent exotic in POP7. These results agree with the observations in a similar study conducted on maize (*Zea mays* L.) using various levels of exotic germplasm introgression, where they observed no significant difference between adapted parents and populations with one dose of exotic germplasm (Crossa and Gardner, 1987). Results from this study suggests that

using exotic parents which are similar in flowering cycle with adapted parents could improve genetic base of adapted cultivars without altering days to first flower.

2.3.2 Bolls plant^{-1}:

Test for fixed effects revealed that population, and population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on bolls plant^{-1} (Table 5). Exotic percentage did not have a significant effect on bolls plant^{-1} ($P=0.3758$). Covariance parameter estimates indicate that the nested effect replication within year was not significant. Interaction effects (exotic percentage × replication within year), and (population × exotic percentage × replication within year) were also non-significant. Interaction effect population × replication within year was highly significant at $P \leq 0.0065$, however, the major portion of this interaction may be due to population effect as replication within year was not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 6). Except in population POP2, increase in exotic germplasm up to 50 percent did not show any significant difference. Increase in exotic germplasm beyond 50 percent showed significant decline in bolls plant^{-1} among 5 of 8 populations (POP1, POP2, POP3, POP4 and POP7). It is worth mentioning that the exotic parents were not statistically different from adapted parents except in populations POP3, POP6 and POP7.

A graphical illustration of boll number means clearly indicates a noticeable variation among populations derived from the two exotic parents (Figure 4). In the year 2009, the part of the field where populations POP1, POP2, POP3 and POP4 were planted had a severe infestation

of nematodes. Upon further investigation it was known that the same field was used for nematode trials two years before the testing in 2009. In the year 2010, there was a gradient of shade due to tree line near the experimental plots that might have affected bolls plant^{-1}. The above-mentioned reasons may explain the differences between the same controls present in POP1 to POP4 and POP5 to POP8. However, as each population was treated as a separate experiment and treated similarly, differences within each population should still provide valid information.

From these results it can be inferred that an increase in exotic germplasm introgression may be permissible up to 50 percent without adversely affecting bolls plant^{-1}, but further increase in exotic percentage may lead to decrease in boll number in most cases. In John cotton germplasm lines, which were developed using exotic introgression, some introgressed lines recorded similar boll number as of 2 cultivar checks (Zeng and Meredith, Jr., 2009b). It may be worthwhile to choose exotic parents with higher bolls plant^{-1} as this trait has been reported as moderately heritable (Desalegn et al., 2009). Our scope of inference may be limited to populations derived from exotic parents, which are not significantly different from adapted parents with respect to bolls plant^{-1}.

2.3.3. Boll size:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on boll size (Table 7). Covariance parameter estimates indicate that the nested effect (replication within environment) and interaction effect (population × replication within environment) were significant and highly significant respectively. Significant interaction between population and environment indicate

that population performance may have been affected by environment, however the high F value for population effect suggest that populations ranks may not likely change in different environments. Interaction effects (exotic percentage × replication within environment), and (population × exotic percentage × replication within environment) were not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 8). An increase in exotic parentage percentage significantly decreased boll size in 5 out of 8 populations (POP1, POP5, POP6, POP7 and POP8). In populations POP2, POP3 and POP4 there was no significant difference observed between exotic percentage groups and their respective controls. Results observed in POP2, POP3, POP4 are in agreement to the results observed in exotic introgression studies where the average boll size of introgressed lines are not significantly different from cultivar checks (Zeng and Meredith, Jr., 2009a, 2009b). In our studies, the decline in boll size was more noticeable in populations derived using TX1419 as exotic parent than the populations derived using exotic parent TX245.

Graphical illustration of boll size means shows consistency in the mean performance of adapted parents across different experiments (Figure 5). Means of exotic parents also showed consistency across experiments. Populations developed using TX245 as exotic parent had relatively bigger bolls compared to populations developed using TX1419 as exotic parent. It is also noticeable that boll size dropped up to one gram in POP5 and POP8 with an increase in exotic germplasm up to 25 percent. The decline in boll size was more pronounced in populations derived from exotic parent TX1419, which could be due to the fact that TX1419 had smaller bolls than the adapted parents to begin with.

From these results it can be inferred that the boll size significantly declined with an increase in exotic percentage among populations where exotic parent was significantly different from adapted parent, which is clearly evident from this study. The non-significant differences observed in POP2, POP3 and POP4 may be due to the fact that exotic parent not being significantly different from the adapted parents. In 50 percent of the populations boll size was not affected up to 25 percent exotic germplasm introgression. Since the boll sizes of exotic parents were significantly smaller compared to adapted controls in most of the populations studied, boll sizes must have been affected negatively. With exotic parents having bigger boll size better results may be achieved.

2.3.4 Seeds boll^{-1}:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on seeds boll^{-1} (Table 9). Covariance parameter estimates indicate that the nested effect (replication within environment), interaction effects (population × replication within environment) and (exotic percentage × replication within environment) were significant. Interaction effect population × exotic percentage × replication within environment was not significant. These results suggest that our populations were not stable across environments for seeds boll^{-1}.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 10). Differential response was observed among populations derived from TX245 and TX1419 with increase in exotic percentage. An increase in exotic percentage did not show any significant difference with their

respective controls in POP1, POP2 and POP4, which is similar to observations in JC lines where average of introgressed lines was not significantly different from cultivar checks (Zeng and Meredith, Jr., 2009b). In population POP3, an increase in exotic percentage significantly increased seeds boll^{-1}. Populations POP5 and POP8 showed a highly significant decrease in seeds boll^{-1} with an increase in exotic percentage, which is similar to the observations in a species polycross study where some exotic introgressed lines had significantly lower seeds boll^{-1} than cultivar checks (Zeng and Meredith, Jr., 2009a). Populations POP6 and POP7 did not show significant difference up to 25 percent, but further increase in exotic germplasm percentage showed a highly significant decline in seeds boll^{-1}.

Graphical illustration of seeds boll^{-1} means show consistency in means of adapted parents across experiments (Figure 6). Means of exotic parents also seemed consistent across experiments. Populations (POP1-4) developed using TX245 as exotic parent appeared to be more or less similar to adapted controls except in POP3 where exotic parent was better than the adapted parent. Populations (POP5-8) developed using TX1419 as exotic parent showed almost linear decrease with an increase in exotic percentage. The differential response observed among populations derived from two exotic parents may be due to the fact that TX245 has more seeds boll^{-1} compared to adapted parents, whereas TX1419 had less seeds boll^{-1} than adapted parents.

Based on these results it can be inferred that an increase in exotic percentage may significantly decrease seeds boll^{-1} if the exotic parent has significantly lower seeds boll^{-1} than the adapted parent. However, an increase in exotic percentage up to 25 percent did not show significant decline in 6 out of 8 populations, which indicates there is a high probability of expanding the genetic base of cotton without a reduction in seeds boll^{-1}. Non-significant differences observed in populations POP1, POP2 and POP4 can be attributed to the exotic parent

being not significantly different from adapted cultivars. The significant improvement observed in population POP3 can be attributed to the exotic parent being superior to the adapted parent in this population, whereas, the highly significant decline observed in populations POP5-8 could be attributed to the exotic parent having a lower seeds boll^{-1} than adapted controls. Non-significant differences observed in POP6 and POP7 suggests that this trait can still buffer the exotic germplasm effect up to 25 percent even though exotic parent was inferior.

2.3.5. Lint seed^{-1}:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had highly significant effects ($P \leq 0.0001$) on lint seed^{-1} (Table 11). Covariance parameter estimates indicate that replication within environment was significant at $P \leq 0.05$, which is on the verge of being non-significant. Interactions (population × replication within environment) and (exotic percentage × replication within environment) were highly significant. In the interactions (population × replication within environment) and (exotic percentage × replication within environment) major portion of the variation may likely be due to population and exotic percentage effects respectively, since replication within environment was almost close to being non-significant. The population × exotic percentage × replication within environment interaction was not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 12). Except in populations POP6 and POP7, an increase in exotic percentage significantly decreased lint seed^{-1} in all populations in this study. In population POP6, exotic percentage introgression at 25 and 75

percent exotic did not show any significant difference from the adapted parent, however, a highly significant difference was observed between 50 and 100 percent exotic compared to the adapted parent. In population POP7 no significant difference was observed up to 25 percent exotic percentage increase, but further increase resulted in a highly significant decline in lint seed^{-1}.

Graphical illustration of means showed consistency of adapted parent controls across experiments (Figure 7). Exotic parents also show consistency across experiments. The decline in lint seed^{-1} with an increase in exotic percentage was more severe in populations derived from exotic parent TX245 than populations developed from TX1419. Although both exotic parents are significantly inferior to adapted parents, the magnitude of the negative impact appears to be much higher in populations derived from exotic parent TX245, suggesting that the impact of exotic germplasm was highly dependent upon the population and that response varied widely.

From these results it can be concluded that increase in exotic germplasm adversely affects lint seed^{-1}. Since both the exotic parents used this study were significantly inferior to adapted parents used, this negative decline can be logically expected. Inferior lint seed^{-1} in exotic germplasm introgressed lines when compared to cultivars checks was also reported in another introgression study (Zeng and Meredith, Jr., 2009b). Non-significant differences observed in populations POP6 and POP7 offers some promise, but this lack of differences could also be due to chance. Using exotic parents, which are equivalent or better in lint seed^{-1} may show different results.

2.3.6 Seed cotton yield:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on seed cotton yield

(Table 13). Covariance parameter estimates indicate that the nested effect replication within environment was not significant. Interaction (population × replication within environment) was highly significant ($P \leq 0.0039$), whereas interactions (exotic percentage × replication within environment) and (population × exotic percentage × replication within environment) were significant at $P \leq 0.0375$ and $P \leq 0.0268$ respectively. The major portion of these interactions may be due to the effects of population, and exotic percentage as the replication within environment interaction was not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 14). An increase in exotic percentage up to 25 percent did not significantly affect seed cotton yields in 6 out of 8 populations; POP1, POP2, POP3, POP6, POP7 and POP8. An increase in exotic percentage beyond 25 percent resulted in significant reduction in seed cotton yield among all the populations with the exception of POP3, which showed a non-significant difference at that level.

Graphical illustration of seed cotton yield means showed consistency of adapted controls across experiments (Figure 8). In the populations derived using TX245 as exotic parent, seed cotton yield response to increase in exotic percentage beyond 25 percent appeared to be almost a linear decline. In the populations derived using TX1419 as exotic parent increase in exotic percentage beyond 25 percent did not show any consistent pattern. Exotic parents TX245 and TX1419 were significantly inferior to adapted controls in all populations, but the decline in seed cotton yield was more drastic in populations derived from exotic parent TX245.

Based on these results we conclude that an increase in exotic germplasm introgression up to 25 percent may have a limited effect on seed cotton yield under the conditions of this experiment with a limited number of observations. Even though yield is measured somewhat differently in cotton and maize, a similar observation was reported in another exotic germplasm introgression study where they concluded that one generation of backcrossing was enough to increase yield to same level as that of adapted material (Crossa and Gardner, 1987). An increase in exotic percentage beyond 25 percent will almost certainly lead to a significant decline in seed cotton yield. This observation parallels our observations for the effect of exotic germplasm introgression on seeds per boll. Since seed cotton yield is a function of number of bolls, seeds per boll and lint percentage, it is very likely that seeds per boll may have been a key contributor to the non-significant difference observed between the adapted parents and the 25 percent exotic germplasm group (Khan et al., 2010). A key interesting observation in this study is that even though both exotic parents are significantly inferior to all adapted parents, seed cotton yield did not get affected in most cases up to 25 percent exotic germplasm increase.

2.3.7 Lint percentage:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on lint percentage (Table 15). Covariance parameter estimates indicate that the nested effect (replication within environment), and interaction effect (population × exotic percentage × replication within environment) were not significant. Interactions (population × replication within environment) and (exotic percentage × replication within environment) were highly significant. Significant interactions between genotype × environment indicates genotypes performed differently across environments, however major portion of these variation may be due to genotypes as the

replication with environment was not significant. Significant genotype and genotype × environment interactions were also observed exotic introgression studies (Zeng et al., 2007, 2010; Zeng and Meredith, Jr, 2009b)

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 16). In populations POP1, POP2, POP4, POP5, and POP8, a highly significant decline in lint percentage was observed with an increase in exotic parentage percentage. In populations POP3 and POP7, no significant difference was observed up to 25 percent, but further increase resulted in a highly significant decline in lint percentage. In POP6, no significant difference was observed at 25 and 75 percent, but highly significant negative values were observed at 50 percent and 100 percent.

Graphical illustration of LS means for lint percentage indicates consistent performance of adapted parent controls across experiments (Figure 9). Exotic parents also show consistency among experiments in POP1-4 for TX245 and POP5-8 for TX1419. Populations derived from exotic parent TX245 showed an almost linear decline in lint percent, whereas no pattern was observed among populations derived from exotic parent TX1419. Exotic parents TX245 and TX1419 were significantly inferior to adapted controls in all populations, but the decline in lint percentage was more drastic in populations derived from exotic parent TX245.

From these results we conclude that an increase in exotic germplasm adversely affected lint percentages. Since both the exotic parents used this study had significantly lower lint percentage than the adapted parents used, this negative decline could be explained based on parental performance. Similar results were also reported in another exotic introgression study

where the mean lint percentages were significantly lower compared to cultivars used as checks (Zeng and Meredith, Jr, 2009b). Populations derived from TX245 showed an almost linear decrease in lint percentage compared to the populations derived from TX1419. It may be worth mentioning that populations derived from TX245 showed almost similar number of seeds per boll as of adapted controls, but lint percentages declined significantly, which indicates fiber production per seed was not on par with adapted controls. TX245 in its region of adaptation recorded a lint percentage of 40%, but in our study it performed significantly lower (Weaver et al., 2007). In the populations derived from TX1419 number of seeds per boll declined drastically but the decline in lint percentage was not as large as in populations from TX245. These results may imply that the positive and negative interactions may not have been broken by the introgression of exotic germplasm for this particular trait.

2.3.8 Lint yield:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on lint yield (Table 17). Covariance parameter estimates indicate that the nested effect (replication within environment), and interaction effect (population × exotic percentage × replication within environment) were not significant. Interactions (population × replication within environment) and (exotic percentage × replication within environment) were highly significant. Significant interactions between genotype × environment indicates genotypes performed differently across environments, however major portion of these variation may be due to genotypes as the replication with environment was not significant. Significant genotype and genotype × environment interactions were also observed exotic introgression studies (Zeng et al., 2007, 2010; Zeng and Meredith, Jr., 2009a).

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 18). Increase in exotic percentage up to 25 percent did not show any significant difference from the adapted controls in 5 out of 8 populations; POP1, POP3, POP6, POP7 and POP8. Increase in exotic germplasm beyond 25 percent significantly decreased lint yield in all the populations used in this study. Populations POP2, POP4 and POP5 showed a highly significant decline even with 25 percent of exotic germplasm.

Graphical illustration of lint yield means shows consistent performance of adapted parents across experiments (Figure 10). The exotic parent control TX245 performance seems to be variable across experiments, but the exotic parent TX1419 performed almost consistently in all experiments. Populations derived from TX245 as the exotic parent showed an almost linear decline in lint yield with an increase in exotic percentage, which may have been largely due to decreases in lint seed^{-1} and lint percentage. In three out of four populations derived using TX1419 as the exotic parent an increase in exotic parentage percentage up to 25 percent resulted in no loss in lint yield compared to the adapted parents. However further increase in exotic parent percentage caused a significant drop in lint yield.

Based on these results, it can be inferred that one generation of backcrossing to adapted parent (25% exotic) may lead to a nearly 62 percent success rate in improving genetic base without affecting lint yield, whereas, one generation of backcross to the exotic parent (75% exotic) may lead to 100 percent chance of significant decline in lint yields. An increase in exotic percentage beyond 25 percent led to significant decline in lint yields in almost all populations (McCarty et al., 1996; Tang et al., 1993a). This may be largely due to the major genes affecting

lack of adaptation or favorable genes of exotic germplasm being masked (Crossa and Gardner, 1987). A highly significant decline in lint yield was also reported in another study where the JC introgressed lines recorded a significantly lower mean lint yield compared to the cultivar checks (Zeng and Meredith, Jr., 2009b). The pattern observed with this trait is similar to what we have seen with seed cotton yield, but seed cotton yields were slightly less affected largely due to the role of the seed component in determining the trait. The significant reduction in lint seed^{-1} and lint percentages reflected on lint yields among all the experiments. In populations derived using TX245 as exotic parent, seeds per boll were almost equal to adapted controls in POP1, POP2 and POP4, but that did not reflect in lint yield largely due to a decrease in lint seed^{-1} or lint percentage. Using exotic parents, which may be better yielding or having a better combining ability may help in increasing success rates. However, it may be unlikely to obtain more positive results because of adaptability issues associated with most plant introductions. Results observed in studies using day neutral accessions arrived at similar conclusions (McCarty et al., 2005, 2007).

2.3.9 Lodging:

Tests for fixed effects revealed that population had a significant effect ($P \leq 0.0239$), whereas exotic percentage, and population × exotic percentage had a highly significant effect ($P \leq 0.0033$ and $P \leq 0.0001$ respectively) on lodging (Table 19). Covariance parameter estimates indicate that nested effect (replication within year), and interaction effects (population × replication within year), (exotic percentage × replication within year), and (population × exotic percentage × replication within year) were non significant. The non-significance of these interactions indicates our populations were stable across environments for lodging.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 20). An increase in exotic germplasm did not show any statistically significant difference for lodging in 5 out of 8 populations; POP2, POP3, POP4, POP5, and POP6. In population POP1, an increase in exotic germplasm even of just 25 percent resulted in a statistically significant increase in lodging, while a further increase in exotic percentage had no additional effect. In population POP7, increase in exotic percentage up to 25 percent resulted in improving lodging resistance, however further increases in exotic percentage showed no significant difference. In population POP8, except for 50 percent exotic, increase in exotic percentage resulted in a significant increase in lodging.

Graphical illustration of lodging score means is presented in figure 11. From the graph it is apparent that almost all the data points ranged between lodging score 1 to 3. Except in POP3, exotic parent TX245 performed consistently among all three experiments, whereas exotic parent TX1419 performed almost consistently among all the experiments.

From these results it can be inferred that an increase in exotic percentage did not result in any negative effect on lodging in most cases. It is interesting to see that adapted control FM966 in POP1 and POP5 recorded totally different results with two different exotic parent controls. Although in POP1 and POP8 there was a significant increase in lodging score, plants did not lodge completely. These results agree with a similar study in soybean where they observed a non-significant difference in lodging up to 50 percent exotic germplasm introgression (Ininda et al., 1996).

2.3.10. Earliness:

Tests for fixed effects revealed that population and exotic percentage had a highly significant (P=0.0106) and significant effect (P=0.0344) respectively (Table 21). Interaction population × exotic percentage was not significant. Covariance parameter estimates indicate that all the tested interaction effects were non significant. Since this experiment was conducted in one year and one location only, scope of these results is very limited. Mean comparisons between exotic percentage groups and their respective adapted controls within each population are presented in Table 23. Even though interaction effect was not significant we chose to present mean comparisons within each population as opposed to combined means across populations. Increase in exotic parentage up to 25 percent did not show any significant difference in all the 3 populations, whereas any further increase in exotic percentage resulted in a significant delay in maturity in POP6 and POP7, and a statistically non-significant delay in boll maturity in POP8. Interestingly, further increase in exotic percentage up to 75 percent resulted in a highly significant delay in maturity in POP8, but difference in earliness was observed in other 2 populations; POP6 and POP7.

Graphical illustration of earliness means is presented in Figure 12. From the graph it is apparent that these populations did not follow any predictable trend. Exotic parent TX1419 recorded inconsistent performance across the experiments. The lower mean earliness percentages in POP 7 may be due to misjudgment in first picking time. Results from this limited data indicate that an increase in exotic percentage up to 25 percent may not negatively impact earliness. Further increases in exotic percentage may be unpredictable. The data is very limited so the scope of these results by necessity is very limited also.

Table 3. ANOVA results and covariance parameter estimates for days to first flower.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	21	2.66*	0.0387
Exotic Percentage	4	12	6.65**	0.0037
Population × Exotic Percentage	28	84	3.86**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Year)	13.78	11.39	1.21	0.1132
Pop×Rep(Year)	0.84**	0.34	2.46	0.0070
ExoPct×Rep(Year)	0.16	0.14	1.14	0.1276
Pop×ExoPct×Rep(Year)	0.43*	0.22	2.00	0.0228

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 4. Least square mean comparisons for days to first flower.

	Days to first flower							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**55.12**	**52.87**	**54.42**	**53.79**	**53.17**	**55.04**	**56.08**	**55.62**
EP1	-0.67	-0.98	-0.87	0.16	-0.72	-1.54	-1.58	-0.98
EP2	-0.47	0.82	-0.12	1.01	-0.12	-1.39	-0.88	-0.58
EP3	1.48	4.22**	0.88	1.26	-1.27	-1.34	-3.23*	-2.08
EP4	0.69	3.50*	2.02	0.90	-0.85	-1.79	-1.71	-1.44
SE†	1.10	1.46	1.30	0.95	0.66	1.36	1.30	1.31

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.
† Standard error
* Significant at P ≤0.05
** Significant at P ≤0.01

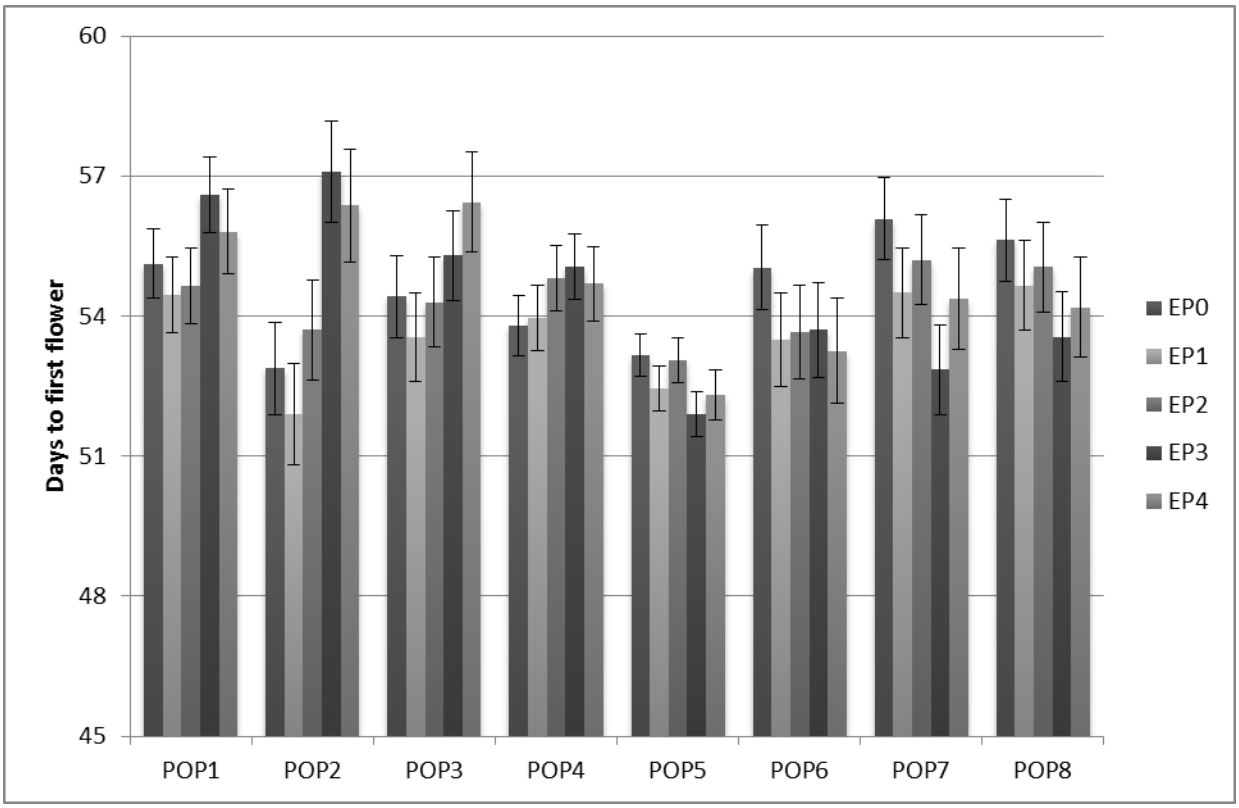

Figure 3. Effect of exotic germplasm introgression on days to first flower. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 5. ANOVA results and covariance parameter estimates for boll plant^{-1}.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	21	11.00**	≤0.0001
Exotic Percentage	4	12	1.16	0.3758
Population × Exotic Percentage	28	84	3.58**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Year)	-0.05	0.27	-0.18	0.8601
Pop×Rep(Year)	1.98**	0.73	2.72	0.0065
ExoPct×Rep(Year)	0.30	0.25	1.21	0.2266
Pop×ExoPct×Rep(Year)	-0.47	0.31	-1.55	0.1219

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 6. Least square mean comparisons for boll plant^{-1}.

	Boll plant^{-1}							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**7.77**	**7.45**	**8.27**	**7.36**	**9.52**	**10.59**	**9.77**	**9.41**
EP1	-1.00	-0.01	-0.95	-0.48	0.20	-1.58	2.11	-1.33
EP2	-1.21	-1.83*	-0.75	-1.60	0.35	1.89	-0.42	-0.37
EP3	-2.38**	-2.23**	-2.55**	-2.06*	0.64	1.71	2.95*	0.91
EP4	-1.15	0.11	-2.85**	-1.43	-0.20	3.55*	3.03*	2.09
SE†	0.71	0.74	0.83	0.88	1.16	1.49	1.18	1.19

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

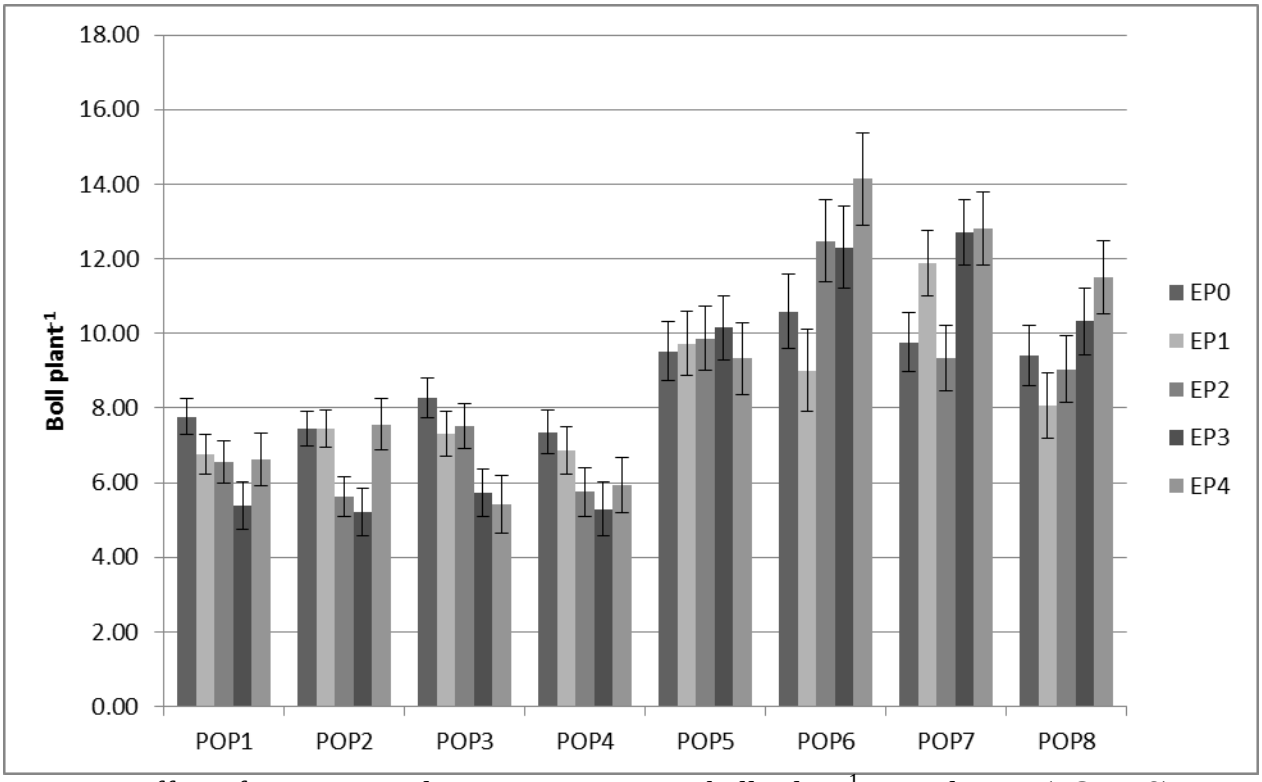

Figure 4. Effect of exotic germplasm introgression on bolls plant^{-1}. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 7. ANOVA results and covariance parameter estimates for boll size.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	49	32.93**	≤0.0001
Exotic Percentage	4	28	41.99**	≤0.0001
Population × Exotic Percentage	28	196	11.05**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.51	0.28	1.83	0.0333
Pop×Rep(Environment)	0.02**	0.01	3.33	0.0004
ExoPct×Rep(Environment)	0.00	0.00	1.06	0.1435
Pop×ExoPct×Rep(Environment)	0.00	0.01	0.11	0.4568

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 8. Least square mean comparisons for boll size.

	Boll size(g)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**5.50**	**5.07**	**4.46**	**4.78**	**5.39**	**4.90**	**4.40**	**4.88**
EP1	-0.54*	0.13	0.15	-0.12	-0.91**	-0.03	-0.38*	-0.85**
EP2	-0.53*	-0.24	0.09	-0.19	-1.28**	-1.06**	-0.82**	-0.97**
EP3	-0.70**	-0.22	0.21	-0.16	-1.23**	-0.55**	-0.66**	-1.08**
EP4	-1.06**	-0.10	0.05	-0.11	-1.70**	-1.08**	-0.77**	-0.99**
SE†	0.22	0.19	0.18	0.17	0.19	0.18	0.18	0.18

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

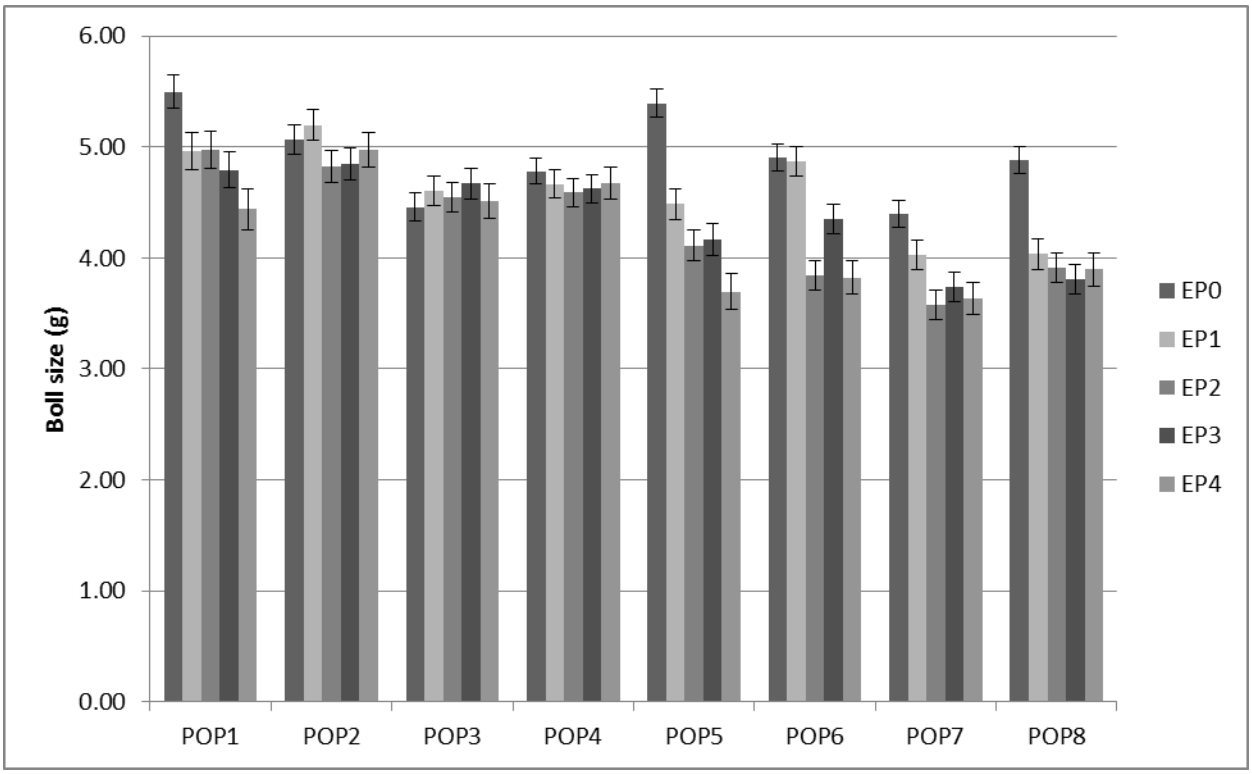

Figure 5. Effect of exotic germplasm introgression on boll size (g). Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 9. ANOVA results and covariance parameter estimates for seeds boll^{-1}.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	49	28.75**	≤0.0001
Exotic Percentage	4	28	10.30**	≤0.0001
Population × Exotic Percentage	28	196	11.68**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	13.89*	7.75	1.79	0.0365
Pop×Rep(Environment)	1.61**	0.35	3.87	≤0.0001
ExoPct×Rep(Environment)	0.23*	0.13	1.75	0.0399
Pop×ExoPct×Rep(Environment)	0.23	0.20	1.12	0.1315

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 10. Least square mean comparisons for seeds boll^{-1}.

	Seeds boll^{-1}							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**31.00**	**30.78**	**28.79**	**29.81**	**30.26**	**30.89**	**27.00**	**30.72**
EP1	-0.32	0.92	2.65*	0.48	-2.67**	0.38	-1.59	-4.58**
EP2	0.60	1.47	2.20*	0.46	-4.83**	-5.21**	-2.84**	-5.29**
EP3	-0.43	1.09	2.55*	1.45	-5.08**	-3.55**	-3.14**	-6.54**
EP4	-1.11	0.80	2.45*	0.98	-7.09**	-6.86**	-4.14**	-4.46**
SE†	1.33	1.05	1.17	1.02	0.96	1.02	1.01	1.10

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

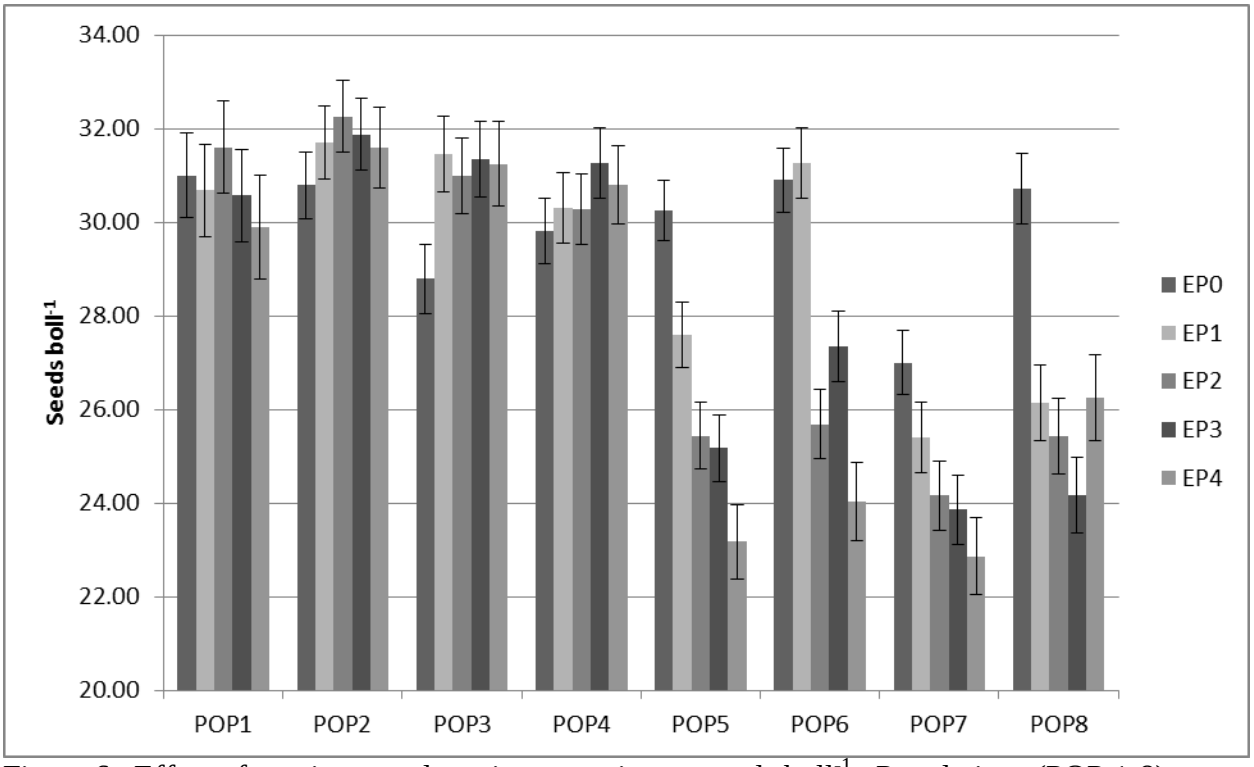

Figure 6. Effect of exotic germplasm introgression on seeds boll^{-1}. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 11. ANOVA results and covariance parameter estimates for lint seed^{-1}.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	49	5.13**	0.0002
Exotic Percentage	4	28	62.39**	≤0.0001
Population × Exotic Percentage	28	196	9.93**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.16*	0.09	1.69	0.0452
Pop×Rep(Environment)	0.08**	0.02	4.19	≤0.0001
ExoPct×Rep(Environment)	0.02**	0.01	2.49	0.0064
Pop×ExoPct×Rep(Environment)	0.01	0.01	1.32	0.0939

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 12. Least square mean comparisons for lint seed^{-1}.

	Lint seed^{-1} (g)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**7.24**	**7.03**	**6.36**	**7.14**	**7.42**	**6.65**	**6.70**	**6.78**
EP1	-0.95**	-0.44**	-0.50**	-1.10**	-1.34**	-0.08	-0.27	-0.58*
EP2	-1.51**	-1.68**	-0.89**	-1.37**	-1.08**	-1.02**	-1.06**	-0.73**
EP3	-1.61**	-1.90**	-0.97**	-1.59**	-0.75**	-0.12	-0.86**	-0.88**
EP4	-2.13**	-1.58**	-1.33**	-1.78**	-1.62**	-0.88**	-0.74**	-1.05**
SE†	0.14	0.14	0.15	0.17	0.19	0.14	0.17	0.21

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

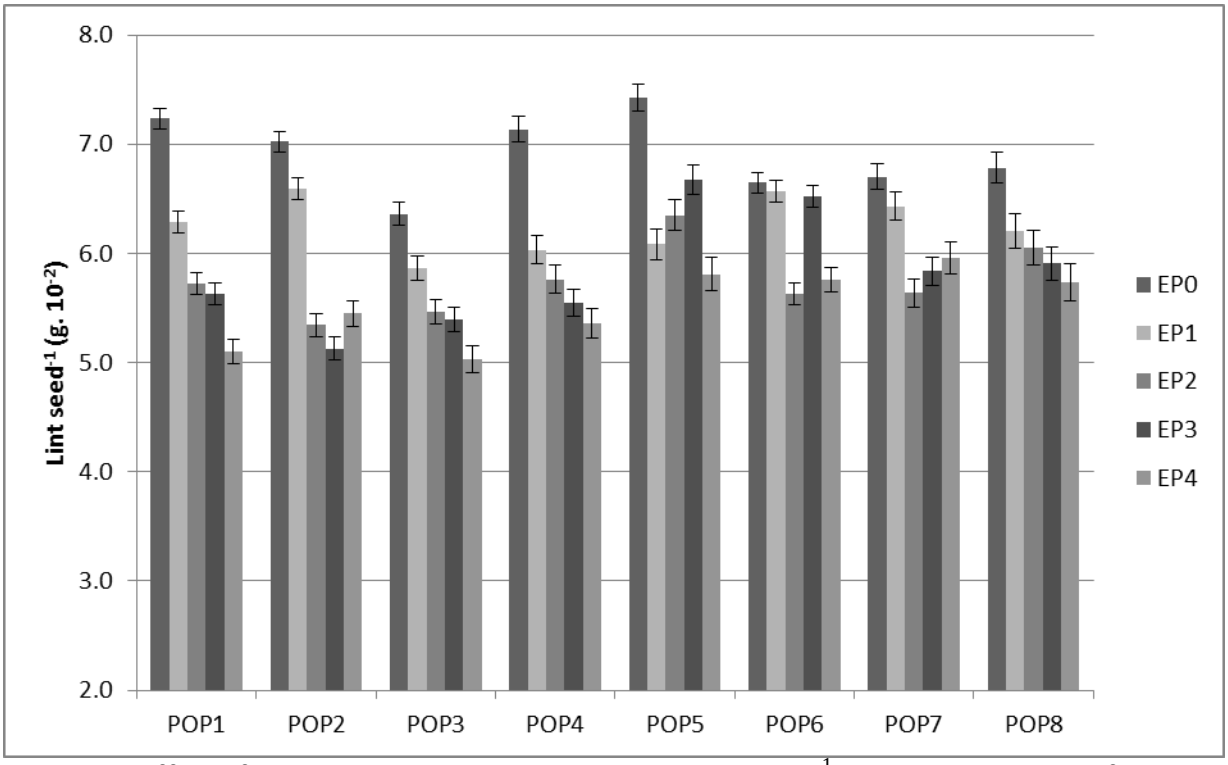

Figure 7. Effect of exotic germplasm introgression on lint seed^{-1} (expressed in grams for 100 seed). Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 13. ANOVA results and covariance parameter estimates for seed cotton yield.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	49	7.35**	≤0.0001
Exotic Percentage	4	28	18.77**	≤0.0001
Population × Exotic Percentage	28	196	5.05**	≤0.0001
Covariance Parameter estimates				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	88681	91839	0.97	0.1671
Pop×Rep(Environment)	66854**	25140	2.66	0.0039
ExoPct×Rep(Environment)	90388*	50758	1.78	0.0375
Pop×ExoPct×Rep(Environment)	15358*	7954	1.93	0.0268

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 14. Least square mean comparisons for seed cotton yield.

	Seed cotton yield (Kg/ha)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**2796**	**3122**	**2509**	**2275**	**2797**	**2991**	**2334**	**2261**
EP1	-74	-296	268	-311*	-558**	33	-103	-6
EP2	-550**	-1037**	-311	-722**	-665**	-930**	-616**	-479*
EP3	-1060**	-1065**	-522**	-1060**	-508*	-753**	-735**	-575**
EP4	-1430**	-1416**	-696**	-1076**	-1054**	-829**	-649**	-459*
SE†	170	178	173	162	197	209	140	187

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

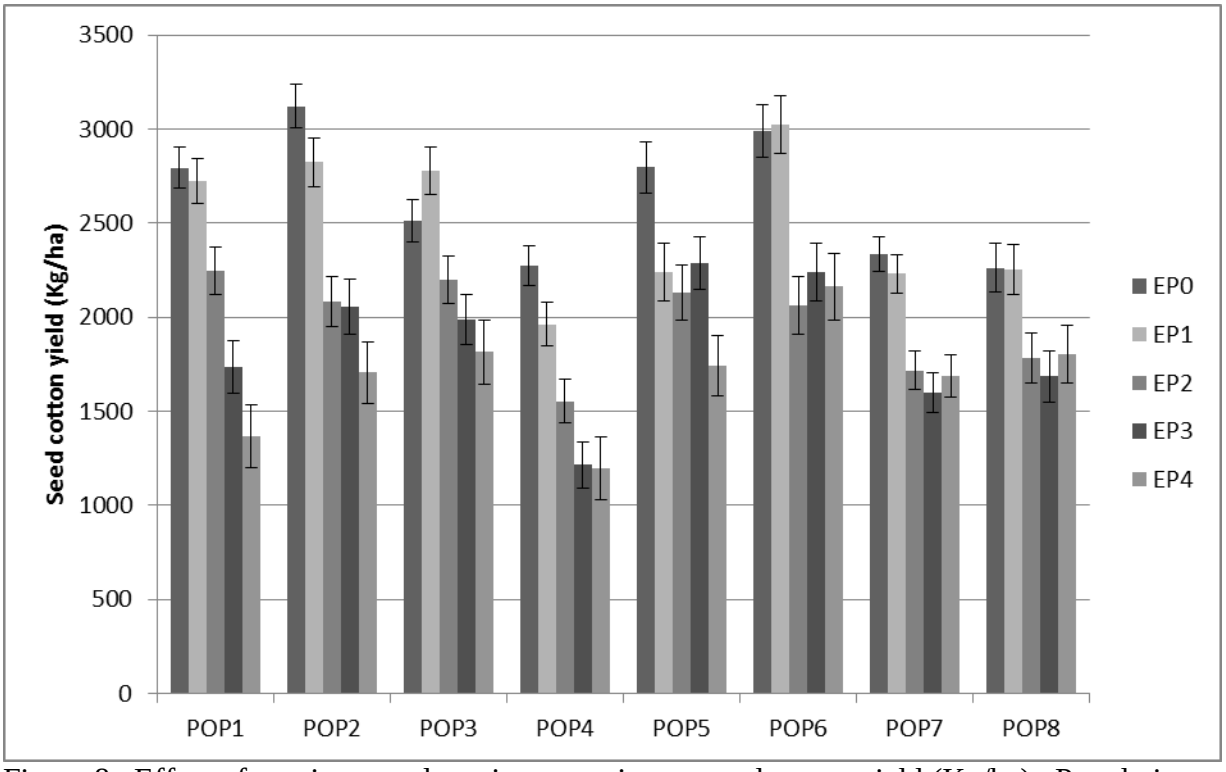

Figure 8. Effect of exotic germplasm introgression on seed cotton yield (Kg/ha). Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 15. ANOVA results and covariance parameter estimates for lint percentage.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	49	8.01**	≤0.0001
Exotic Percentage	4	28	61.95**	≤0.0001
Population × Exotic Percentage	28	196	15.73**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.30	0.29	1.04	0.1493
Pop×Rep(Environment)	0.67**	0.18	3.81	≤0.0001
ExoPct×Rep(Environment)	0.55**	0.18	3.09	0.0010
Pop×ExoPct×Rep(Environment)	0.03	0.09	0.39	0.3483

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 16. Least square mean comparisons for lint percentage.

	Lint percentage (%)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**41.05**	**42.99**	**41.22**	**44.67**	**41.84**	**42.22**	**41.32**	**42.67**
EP1	-2.03**	-2.53**	-1.17	-5.45**	-4.26**	0.38	-0.66	-2.61**
EP2	-4.17**	-7.07**	-3.96**	-6.40**	-2.36**	-4.43**	-3.14**	-3.24**
EP3	-5.00**	-9.04**	-4.67**	-7.01**	-1.40**	-0.89	-4.03**	-5.35**
EP4	-6.75**	-8.14**	-6.10**	-9.27**	-4.99**	-5.77**	-3.55**	-3.77**
SE†	0.53	0.46	0.61	0.58	0.51	0.51	0.57	0.70

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

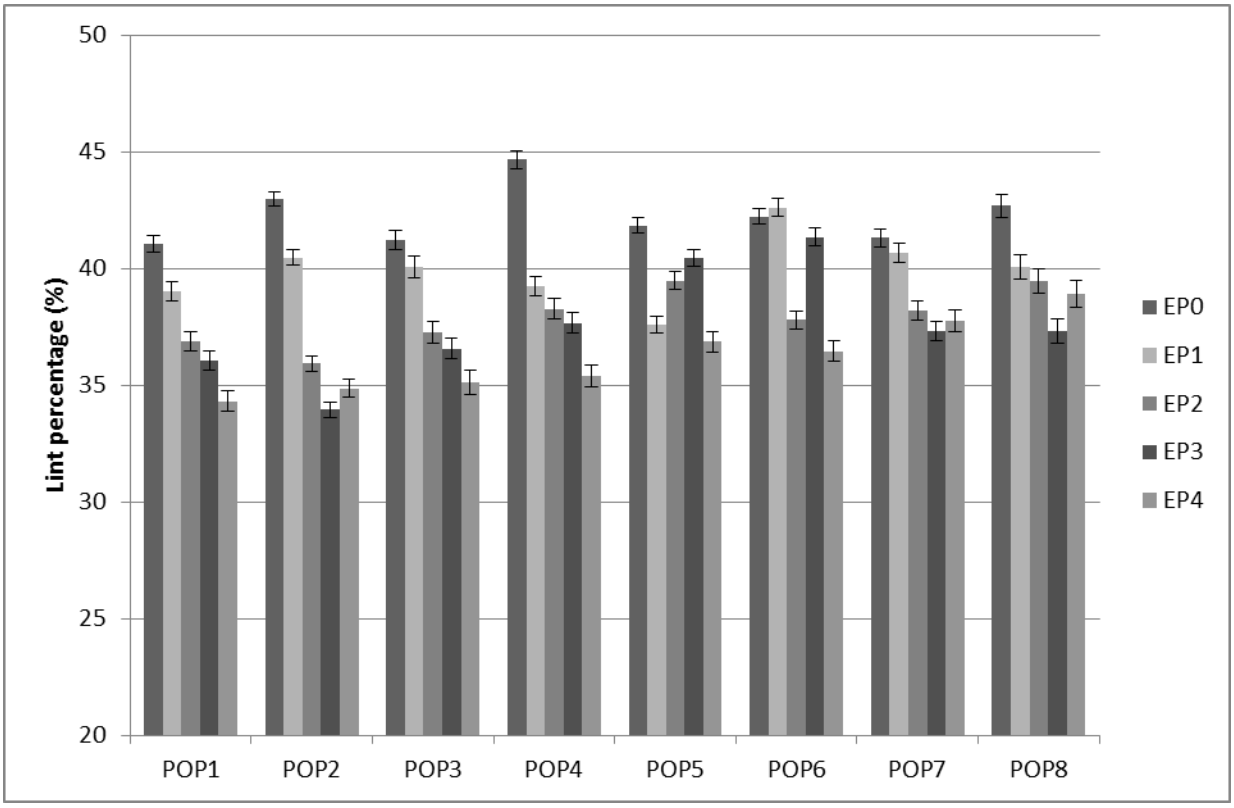

Figure 9. Effect of exotic germplasm introgression on lint percentage (%). Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 17. ANOVA results and covariance parameter estimates for lint yield.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	49	6.81**	≤0.0001
Exotic Percentage	4	28	22.30**	≤0.0001
Population × Exotic Percentage	28	196	7.24**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	3078	4022	0.77	0.2220
Pop×Rep(Environment)	9408**	2377	3.96	≤0.0001
ExoPct×Rep(Environment)	12127**	3602	3.37	0.0004
Pop×ExoPct×Rep(Environment)	1214	1254	0.97	0.1663

* Significant at P ≤0.05

** Significant at P ≤0.01

Table 18. Least square mean comparisons for lint yield.

	Lint yield (Kg/ha)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**1149**	**1339**	**1034**	**1023**	**1182**	**1272**	**970**	**977**
EP1	-87	-200**	86	-244**	-338**	24	-58	-73
EP2	-319**	-591**	-220**	-427**	-346**	-486**	-313**	-269**
EP3	-510**	-636**	-304**	-569**	-251**	-343**	-373**	-345**
EP4	-676**	-739**	-378**	-583**	-539**	-488**	-328**	-271**
SE†	68	72	70	69	82	90	60	82

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

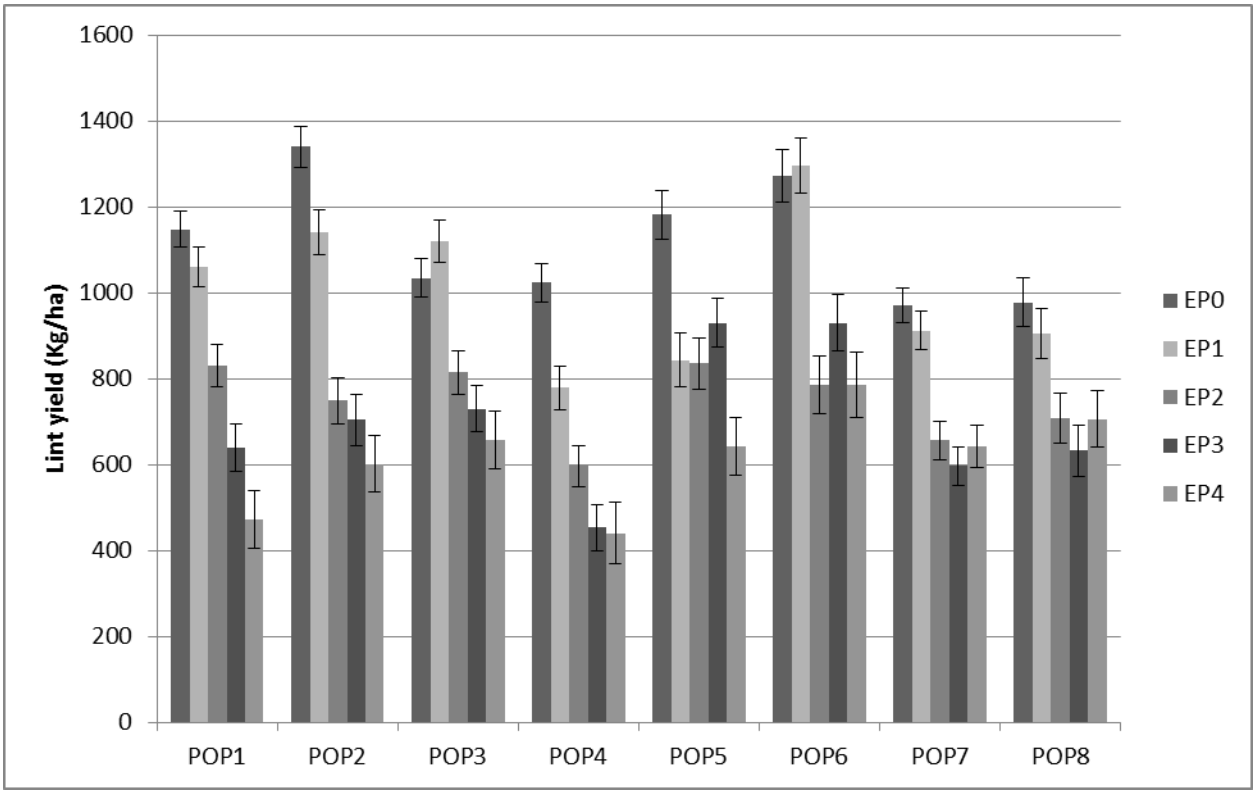

Figure 10. Effect of exotic germplasm introgression on lint yield (Kg/ha). Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 19. ANOVA results and covariance parameter estimates for lodging.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	21	3.00*	0.0239
Exotic Percentage	4	12	7.24**	0.0033
Population × Exotic Percentage	28	84	3.50**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Year)	0.00	0.01	0.43	0.6691
Pop×Rep(Year)	0.01	0.01	1.35	0.1765
ExoPct×Rep(Year)	0.00	0.00	1.01	0.3111
Pop×ExoPct×Rep(Year)	0.00	0.00	1.05	0.2946

* Significant at P ≤0.05
** Significant at P ≤0.01

Table 20. Least square mean comparisons for lodging.

	Lodging							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**1.56**	**2.37**	**2.28**	**2.44**	**1.77**	**2.14**	**2.30**	**2.04**
EP1	0.64**	0.08	-0.23	0.16	0.23	0.11	-0.45*	0.41*
EP2	0.69**	-0.12	0.02	-0.04	0.08	0.31	-0.35	-0.14
EP3	0.69**	-0.12	-0.13	0.01	0.33	0.11	0.05	0.61**
EP4	0.78**	0.12	-0.34	0.21	0.82**	0.57**	0.66**	0.88**
SE†	0.18	0.18	0.18	0.18	0.18	0.18	0.18	0.18

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

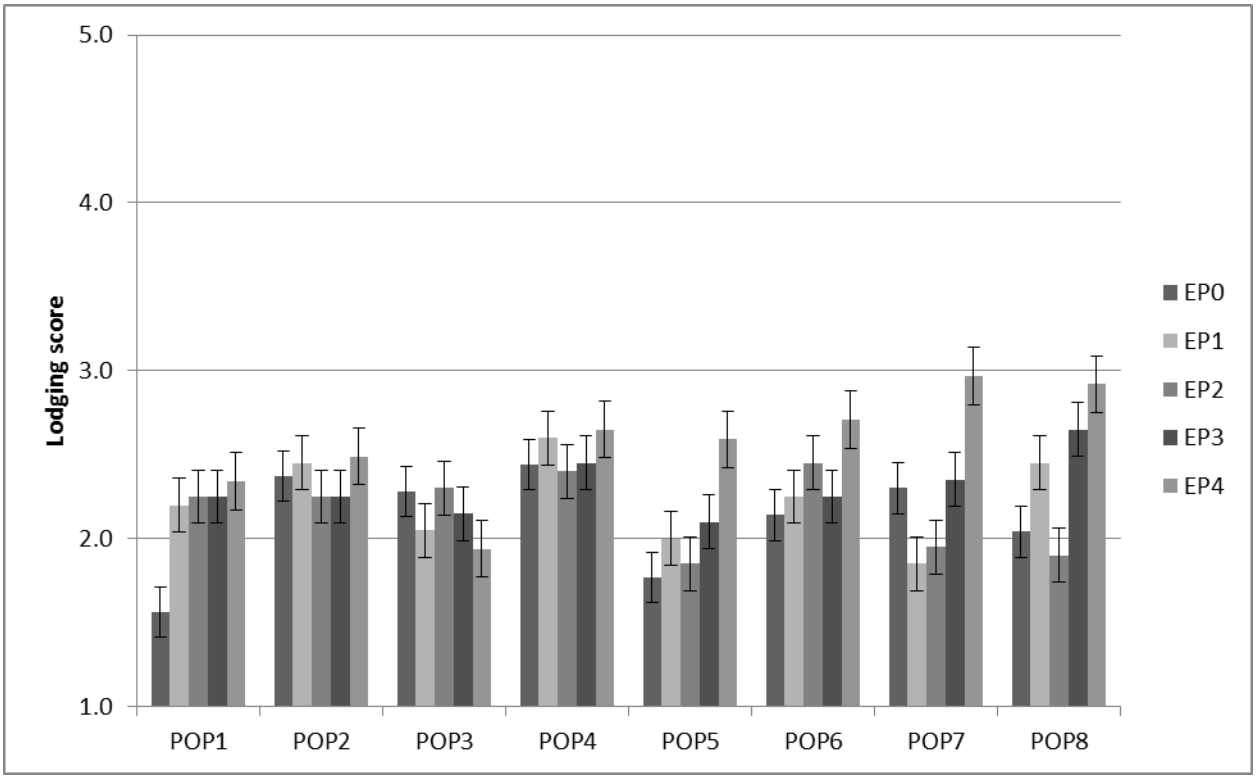

Figure 11. Effect of exotic germplasm introgression on lodging. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)). Lodging score ($1=90^0$, $2=67.5^0$, $3=45^0$, $4=27.5^0$ and $5=0^0$ angle of inclination)

Table 21. ANOVA and covariance parameter estimates for earliness.

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	2	3	29.47**	0.0106
Exotic Percentage	4	4	7.98*	0.0344
Population × Exotic Percentage	8	8	1.87	0.1956
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep	0.03	0.12	0.28	0.7824
Rep×Pop	0.06	0.15	0.42	0.6720
Rep×Exopct	-0.06	0.06	-1.01	0.3129
Rep×Pop×ExoPct	-0.19	0.17	-1.14	0.2527

* Significant at P ≤0.05
** Significant at P ≤0.01

Table 22. Least square mean comparisons for earliness.

	Earliness (%)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	-	-	-	-	-	**85.2**	**58.6**	**85.84**
EP1	-	-	-	-	-	4.7	-1.3	-9.3
EP2	-	-	-	-	-	-26.1**	-18.8*	-16.3
EP3	-	-	-	-	-	-0.1	-1.6	-23.3**
EP4	-	-	-	-	-	-20.4*	-11.2	-12.9
SE†	-	-	-	-	-	6.9	6.9	6.9

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

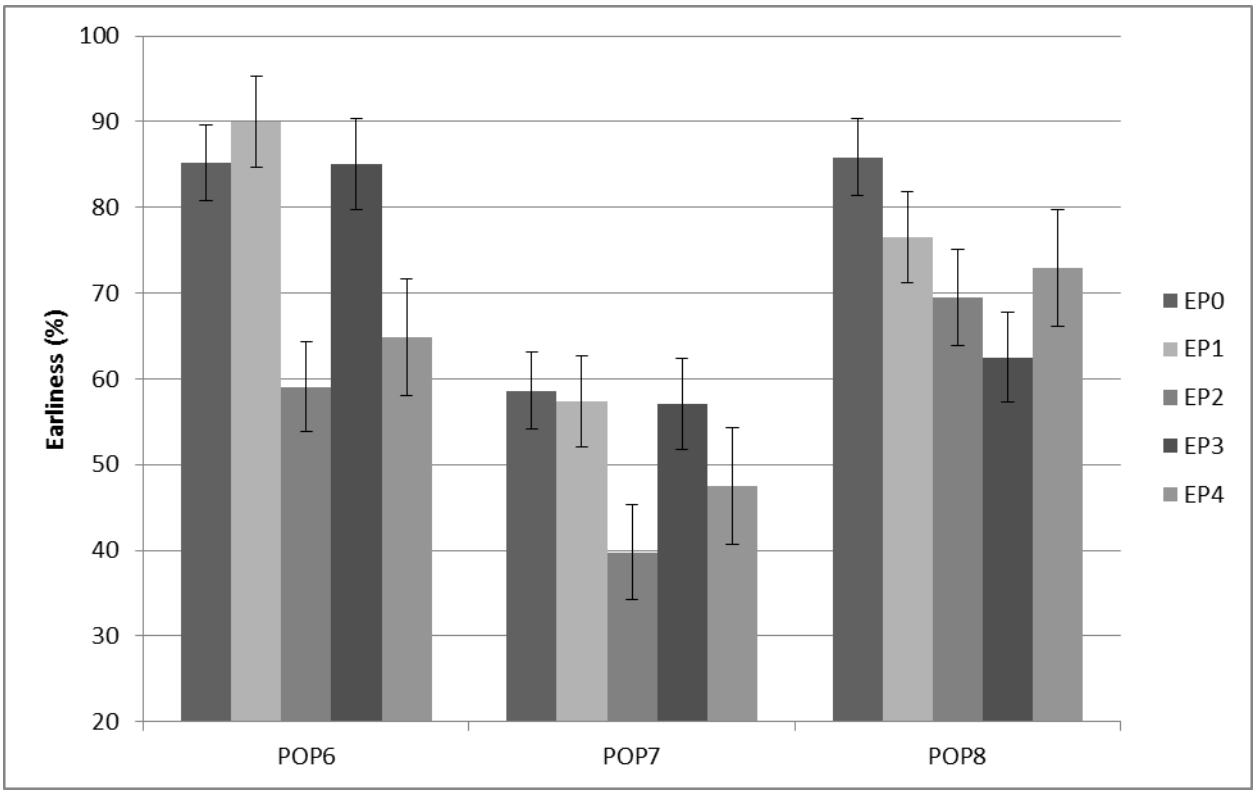

Figure 12. Effect of exotic germplasm introgression on earliness (%). Populations (POP6, POP7 and POP8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

3. Effect of exotic germplasm on fiber properties

ABSTRACT

Upland cotton (*Gossypium hirsutum* L.) is an economically important natural fiber crop in the world. Along with fiber, it is also valued for its oil and protein portion of the seed. Upland cotton is reported to be facing a risk associated with a narrow genetic base. The utilization of closely related parents and many reselections within elite cultivars in the development of cultivars has led to a narrow genetic base. Genetic uniformity predisposes crops to various abiotic and biotic stresses. Cotton cultivars with increased genetic diversity can offer plasticity to be able to respond to stressful environments. Exploration of exotic germplasm for introgressing novel genes and broadening the genetic base of cotton may also offer additional genetic variation to help future trait improvement. Our research is focused on the objective of determining the effect of exotic germplasm on fiber properties of adapted cotton cultivars. Two plant introductions (TX245 and TX1419) were systematically crossed with four cultivars (FM966, PM1218, Deltapearl and SG747) to derive five groups of lines with 0, 25, 50, 75 and 100 percent exotic germplasm. Experimental materials were tested in a randomized complete block design (RCBD) and analyzed using mixed models procedures as implemented in SAS® PROC MIXED (SAS 9.2, SAS Institute, Cary, NC).

Results revealed highly significant effects due to population, exotic percentage, and population × exotic percentage interactions for all the fiber traits. Genotype × environment interactions were also significant for most of the traits. An increase in exotic parentage

percentage significantly lowered fiber properties, however the magnitude of effect varied with populations and exotic parentage percentage levels. Among fiber properties, micronaire was the most affected trait, whereas fiber elongation and short fiber content were the least affected traits with an increase in exotic germplasm introgression. Populations derived from exotic parent TX245 showed a significant reduction in fiber length, fiber strength, micronaire, and fiber uniformity, but fiber elongation improved with an increase in exotic parentage percentage and remained unaffected in most cases for short fiber content. Populations derived from exotic parent TX1419 gave mixed results with an increase in exotic parentage percentage. Except for fiber elongation, fiber properties remained unaffected or improved in POP6 and POP8 with an increase in exotic parentage percentage. Fiber strength improved significantly with an increase in exotic germplasm in POP8. In almost all cases where the exotic parent was better or equal with performance of adapted cultivars, an increase in exotic parentage percentage up to 25 percent did not show a significant difference in fiber properties, which suggests that using exotic parents which are similar or better in fiber properties could expand the genetic base of adapted cultivars without altering fiber properties.

3.2 Materials and methods

This research was conducted in summer 2009 and 2010 at two locations, Tallassee, Alabama and Florence, South Carolina. The first site was located at the Plant Breeding Unit, E.V. Smith Research Center, Tallassee, AL (32º29′N, 85º53′W) and the second site at Coastal Plains Soil, Water and Plant Research Center, Florence, SC (34º29′N, 79º48′W). Soils were Wickham fine sandy loam soil (fine-loamy, mixed, semiactive, thermic Typic Hapludults) at E.V. Smith Research Center, and Goldsboro loamy sand (fine-loamy, siliceous, thermic Aquic Paleudults) at Pee Dee research center. Monthly averages of temperatures and rainfall for both years and locations are graphically represented in Figure 1 and Figure 2 respectively.

The exotic introgression percentage lines used in this study were developed by crossing among two plant introductions and four adapted cultivars. Plant introductions TX245 (PI 165358) and TX1419 (PI 530110) were selected as exotic parents in this study, because they showed moderate levels of resistance to reniform nematode (*Rotylenchulus reniformis* Linford & Oliveria) in an earlier study (USDA, 2006; Weaver et al., 2007). TX245 is an introduction from the state of Guerrero in Mexico and belongs to the race *latifolium*. TX1419 is an introduction from the state of Bahia in Brazil; its race is not yet classified. Four cultivars FM966, PM1218, Deltapearl, and SG747 were used as adapted parents in developing advanced breeding lines. These four cultivars were selected based on their performance and adaptability and represent the elite cotton germplasm for the mid-south and southeastern USA cotton production regions.

Two plant introductions (TX245, TX1419) were systematically crossed with four cultivars (FM966, PM1218, Delta Pearl, SG747) to develop eight populations (Table 1) with five exotic parentage percentage levels within each population. The parents and their hybrid progeny were intermated in different schemes to form five exotic percentage groups EP0, EP1, EP2, EP3, and EP4 with 0%, 25%, 50%, 75%, and 100% PI germplasm respectively. The mating scheme followed was similar to Fehr and Clark (1973). Adapted parents and exotic parents represent the 0 percent and 100 percent exotic groups respectively (Table 2). $F_{2:4}$ lines were used for 50 percent exotic, whereas 25 and 75 percent exotic were represented by $BC_1F_{2:4}$ lines. The backcross to an adapted parent represents lines for 25 percent exotic, and the backcross to an exotic parent represent lines for 75 percent exotic. Crossing work for developing these advanced breeding lines began in the year 2005 and carried to the next generation in 2006. In the year 2007, a total of 1466 lines from all eight populations (2PIs x 4 cultivars) with the above mentioned exotic percentages were planted for generation advance at E.V. Smith Research Center, Tallassee, AL. Out of these 1466 lines, 6 lines (5 lines + 1 additional line for backup) per each exotic group per population were randomly selected and harvested. These selected lines were again planted in 2008 for seed increase and multiplication.

3.2.1 Field experiment:

Field experiments were conducted in 2009 and 2010 at two locations, Tallassee, Alabama, and Florence, South Carolina. For each of the 8 populations, 5 lines representing each exotic percentage group were planted in a randomized complete block design (RCBD) with 2 years, 2 locations, 2 replications and 5 blocks as class factors. The combination of year and location was considered as environment for the purposes of statistical analysis. Plots were double-row with each row measuring 6 m long with 1 m spacing between rows. In 2009, some

plots at Tallassee location were infested with nematodes and *Fusarium* spp due to the presence of inoculum left over from an earlier nematode trial conducted at the same site two years prior to testing. The infested plots were removed from data analysis in order to reduce the noise. The methodology and the data collection steps are described below.

3.2.2 Data collection:

A 50-boll hand-harvested sample was collected from top, middle and bottom portion of the plants from each plot before machine picking. Boll samples were weighed and ginned in laboratory saw gin to determine yield related traits. A sample of 20 to 25 g clean lint from each plot x year x location was sent to Cotton Incorporated, Cary, NC for the determination of fiber properties using High Volume Instrument (HVI) analysis. During 2010, some of the data points from the Alabama location exhibited erratic HVI readings, in order to avoid noise those data points were removed from data analysis.

3.2.2.1 Fiber length:

Fiber length is the mean length of upper 50 percent half of fibers (also referred to as UHM; upper half mean length). It is reported in both 32nds and 100ths of an inch. In a High Volume Instrument (HVI) a sample of cotton would be grasped by a clamp, which is then combed, brushed, straightened and paralleled. This processed cotton (beard) is passed through an optical sensing point and fiber length is recorded (Cotton Incorporated, 2014).

3.2.2.2 Fiber strength:

Fiber strength is the amount of force required to break a bundle of fibers in tex units, where one tex is equal to the weight of 1000 meters of fiber in grams. In the High Volume

Instrument, a beard of cotton sample is clamped in two sets of jaws $1/8^{th}$ inch apart and the force required to break the fibers is measured and recorded in g tex^{-1} (Cotton Incorporated, 2014).

3.2.2.3 Micronaire:

Fiber fineness is measured using different methods, but the most popular method is air flow method. A clean and well-opened fiber sample of specific size (3.24 grams in the US) is compressed into a cylindrical container of fixed dimensions. In a micronaire instrument compressed air is passed through fiber sample at a definite pressure and the air permeability is measured by a flow meter (Cotton Incorporated, 2014). The recorded measurements are expressed in micronaire units.

3.2.2.4 Fiber uniformity:

Fiber uniformity is the ratio between the mean length and the upper half mean length of fibers and expressed in percentage. The mean lengths and upper half mean lengths measured using HVI are used to calculate this trait (Cotton Incorporated, 2014).

3.2.2.5 Fiber elongation:

Fiber elongation (E1) is a measurement of the elasticity (stretch before breaking) of fibers. Fiber elongation is the length that the fibers will stretch before breaking during the measurement of fiber strength and expressed as percentage.

3.2.2.6 Short fiber content:

Short fiber is defined as the proportion of fibers that is shorter than a specified length. A specified length can vary from one geographical region to other but is generally in the range of

0.25 to 1 inch. In the US, short fibers are those which measure less than or equal to one half inch (12.7mm) in length (Bradow and Davidonis, 2000).

3.2.3 Statistical methods:

Data were checked for normality criteria using Statistical Analysis System SAS® PROC UNIVARIATE. Residuals were plotted in histogram and qq plots by selecting student panel option in PROC GLIMMIX module in SAS version 9.2 (SAS® institute, 2008). Mixed models analysis of variance procedures as implemented in SAS® PROC MIXED was used to analyse the data. Population, exotic percentage, and population × exotic percentage were treated as fixed effects, whereas, years, locations, and replications were specified as random effects using the RANDOM statement. The decision to choose either combined analysis or individual analysis of years and locations was chosen depending on the ANOVA results from the analysis. Mean comparisons between exotic percentage groups and their respective controls (EP0; 0 percent exotic) within each population were performed using Dunnett's multiple comparison procedure. Tests of significance for LSMEANS was performed using slicediff option and for covariance parameters using COVTEST.

3.3 Results and Discussion

3.3.1. Fiber length:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on fiber length (Table 23). Covariance parameter estimates indicate that the nested effect replication within environment was significant ($P \leq 0.0403$). Fiber length is generally influenced by environmental conditions, and although a major portion of the variability can be explained by genotypic differences, environment, and genotype × environment interactions also play a significant role in influencing fiber length (Zeng and Meredith, Jr., 2009a, 2009b; Zeng et al., 2010). The covariance estimate of 0.025 for replication within environment may be too low to have a significant impact. All other tested interactions (population × replication within environment), and (exotic percentage × replication within environment) were not significant (Tang et al., 1993b). The non-significant estimates of these interactions indicate that our populations were stable for fiber length across environments, which was also observed in primitive race accessions introgression study (McCarty et al., 2007).

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 24). An increase in exotic parentage percentage up to 25 percent resulted in a highly significant reduction of fiber length in 5 out of 8 populations. In populations POP5, POP6 and POP8 there was no significant

difference between adapted parents and 25 percent exotic parentage group. An increase in exotic parentage percentage beyond 25 percent resulted in a highly significant decline in fiber length in all populations except in POP6 and POP8. In population POP6, an increase in exotic parentage percentage up to 50 percent increased fiber length significantly, while any further increase in exotic parentage percentage did not show any significant difference. In population POP8, an increase in exotic parentage percentage up to 75 percent had no significant effect on fiber length. Results observed in POP8 may be similar to the results in exotic germplasm introgression study where the average of species polycross lines was not significantly different from cultivar checks (Zeng and Meridith, Jr., 2009a, 2009b).

Graphical illustration of fiber length values shows a clear difference between populations derived from TX245 and TX1419 (Figure 13). Decline in fiber length seems to be more drastic in populations derived from the exotic parent TX245. The same adapted parents across different experiments seems to be consistently within a range of 1 mm, and exotic parents TX245 and TX1419 also stayed consistently within a range of 1mm across experiments. Lower fiber lengths observed in populations derived from exotic parent TX245 may be due to the fact that TX245 had a significantly lower fiber length compared to adapted parents.

From these results it can be inferred that an increase in exotic parentage percentage beyond 25 percent almost guarantees a significant decrease in fiber length if the exotic parent is inferior to adapted parents in fiber length. The exotic parent TX245 was inferior to all four adapted parents in fiber length. An increase in exotic parentage percentage up to 25 percent may have a better chance of maintaining fiber length along with expanding the genetic base if the exotic parent is not significantly inferior to adapted parents, which is evident in populations derived from exotic parent TX1419.

3.3.2. Fiber strength:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on fiber strength (Table 25). Covariance parameter estimates indicate nested effect (replication within environment), the interaction effects (exotic percentage × replication within environment) and (population × exotic percentage × replication within environment) were not significant. The non-significance of these interactions indicate that our populations were stable across environments. The population × replication within environment interaction was significant at $P \leq 0.0298$ (Tang et al., 1993b), however, the major portion of this interaction may be due to the population effect as the replication within environment was not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 26). Increase in exotic parentage percentage resulted in a highly significant decrease in fiber strength in 4 out of 8 populations; POP1, POP2, POP3 and POP7. In populations POP4 and POP5, increase in exotic parentage percentage up to 25 percent did not show significant difference in fiber strength, but any further increase in exotic parentage percentage led to a highly significant decline in fiber strength. In population POP6, an increase in exotic parentage percentage did not show significant difference at 25 and 75 percent exotic introgression, but showed a highly significant increase at 50 percent exotic percentage. In population POP8, an increase in exotic parentage percentage led to a highly significant increase in fiber strength. In populations POP6 and POP8, fiber length of exotic parent TX1419 was significantly better than adapted parents. These results

agree with an exotic germplasm study where 3 out of 4 John cotton exotic introgression lines were significantly better for fiber strength than cultivar checks (Zeng et al., 2010).

Graphical illustration of fiber strength values shows a differential pattern among populations derived from TX245 and TX1419 (Figure 14). Decline in fiber strength values seems to be more drastic in populations derived from TX245 compared to TX1419 derived populations. The same adapted controls in different experiments seem to be consistently within a range of 2 g tex^{-1} whereas both exotic parents were consistently within a range of 1 g tex^{-1} across experiments. Exotic parent TX1419 was almost 3 g tex^{-1} higher than exotic parent TX245 in fiber strength. The fact that exotic parent TX1419 was superior to TX245 in fiber strength explains the higher mean performance of various exotic parentage percentages for populations derived from TX1419.

From these results it can be inferred that for fiber strength an increase in exotic parentage percentage is mostly dependent on performance of the exotic parent. Populations derived from the exotic parent TX1419 performed significantly better than populations derived from exotic parent TX245. Significant improvement in fiber strength was observed in populations POP6 and POP8, which indicates that the genetic base of adapted cultivars can be improved along with improving fiber strength if the exotic parent is significantly better in fiber strength.

3.3.3. Micronaire:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on micronaire values (Table 27). Covariance parameter estimates indicate nested effect (replication within environment), interaction effects (population × replication within environment) and (population

× exotic percentage × replication within environment) were significant. These significant interaction effects suggest that there is a significant portion of variability which is influenced by environment and genotype × environment interactions for this trait. Significant environment, and/or genotype × environmental interactions have been reported for micronaire values in many earlier studies (Jones et al., 2014; McCarty et al., 2007; Wu et al., 2010; Zeng et al., 2007, 2010, 2011; Zeng and Meredith, Jr., 2009a, 2009b). The exotic percentage × replication within environment interaction was not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population Table 28. Increase in exotic parentage percentage increased micronaire in all populations derived from the exotic parent TX245, where some of these differences were significant and some highly significant. However, populations derived from the exotic parent TX1419 showed different results. An increase in exotic parentage percentage up to 25 percent did not show significant difference in all populations derived from TX1419 as the exotic parent. Any further increase in exotic parentage percentage resulted in a highly significant increase in micronaire values in populations POP5 and POP7. Population POP8 did not show significant difference up to 50% exotic introgression, whereas population POP6 did not significant difference up to 75% exotic introgression. It is worth mentioning that all adapted parents were significantly lower in micronaire than their exotic counterpart, but micronaire increased significantly even with 25 percent exotic parentage percentage in populations derived from TX245, whereas no significant difference was observed for populations derived from the exotic parent TX1419 up to 25 percent exotic parentage percentage. These results are similar to the species polycross lines and John cotton germplasm

lines studies where introgressed lines were not significantly different from cultivar checks (Zeng and Meredith, Jr., 2009a, 2009b). Graphical illustration of micronaire values shows a similar pattern across all populations even though the magnitudes of differences vary (Figure 15). Micronaire values ranged from 4.5 to 5.5 among populations, and both exotic parents recorded similar micronaire values across experiments.

From these results it can be inferred that response to an increase in exotic parentage percentage is highly population dependent and response varied widely for micronaire values. Using exotic parents that are lower in micronaire than adapted parents may show better results. Although micronaire values were statistically different from adapted parents, most of the populations derived from exotic parent TX1419 and some from TX245 were still within acceptable limits. Increasing the number of testing environments may be required to adequately estimate micronaire as environments and genotype × environment interactions may also play a major role.

3.3.4. Fiber uniformity:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on fiber uniformity (Table 29). Covariance parameter estimates indicate nested effect (replication within environment) and interaction (population × replication within environment) were significant. Since fiber uniformity is the ratio between mean length and upper half mean length of fibers and is expressed in percentage, conditions that affect fiber length would also likely affect fiber uniformity. Further partitioning of analysis indicates replication within location, and replication within year interactions were not significant, but the overall interaction of replication (year ×

location) was significant at P≤0.0425. Interactions (exotic percentage × replication within environment), and (population × exotic percentage × replication within environment) were not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 30). An increase in exotic parentage percentage up to 25 percent and beyond resulted in a highly significant reduction in fiber uniformity in 3 out of 8 populations. In populations POP1, POP4, and POP5 an increase in exotic parentage percentage up to 25 percent did not show a significant difference, but further increases in exotic parentage percentage led to a highly significant decline in fiber uniformity. In populations POP6 and POP8, an increase in exotic parentage percentage did not show any significant difference up to 75% exotic introgression even though the exotic parent was significantly different from respective adapted control in POP6 and not significantly different in POP8.

Graphical illustration of fiber uniformity index values indicate that the exotic parent TX1419 was one percent higher in fiber uniformity index compared to the exotic parent TX245 (Figure 16). Across populations, fiber uniformity index values ranged from 80.5 to 83.5 percent. Fiber uniformity percentages for the same adapted parents across different experiments were consistently within a range of one percent, likewise the same exotic parents in different experiments also performed consistently.

From these results it can be inferred that the response to an increase in exotic percentage may be unpredictable in populations derived from exotic parents, which are significantly inferior

in fiber uniformity. If the exotic parent is not significantly different from adapted control, genetic variability can be improved without adversely effecting fiber uniformity. This was evident in POP8, and also in POP6, which was not much different from the adapted control.

3.3.5. Fiber elongation:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect ($P \leq 0.0001$) on fiber elongation (Table 31). Covariance parameter estimates indicate that the nested effect (replication within environment), interaction effects (exotic percentage × replication within environment) and (population × exotic percentage × replication within environment) were non-significant; however, the population × replication within environment interaction was significant at $P \leq 0.0101$. Since replication within environment was not significant, variability due to populations may be the major source of variation in the interaction population × replication within environment, which suggests performance of populations may not likely change as the main effect population has a much stronger genotypic value (Ng et al., 2014).

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 32). An increase in exotic parentage percentage significantly increased fiber elongation in 5 out of 8 populations. In POP4, an increase in exotic parentage percentage had no significant effect on fiber elongation irrespective of the level of introgression. In population POP6, increase in exotic parentage percentage up to 50 percent did not show significant difference from the adapted control, but further increase led to a highly significant decrease in fiber elongation. In population POP8,

increase in exotic parentage percentage resulted in a highly significant decline in fiber elongation percentages. The significant improvement in fiber elongation percentages agree with the results in John cotton germplasm lines and species polycross studies where some exotic introgressed lines were significantly better than cultivar checks (Zeng and Meredith, Jr., 2009a, 2009b; Zeng et al., 2010).

Graphical illustration of fiber elongation percentages indicates almost all populations followed an increasing trend except in 3 populations; POP4, POP6, and POP8 (Figure 17). Populations derived from the exotic parent TX245 appear to have improved fiber elongation percentages compared to populations derived using TX1419 as exotic parent. It may be due to the fact that the percent fiber elongation in exotic parent TX245 was significantly better than 3 out of 4 adapted parents used in this study. Exotic parent TX1419 was significantly better in percent fiber elongation in POP5 and POP7, and significantly lower in POP6 and POP8. Overall, fiber elongation percentages among populations ranged from 4 percent to almost 7 percent which is very large in terms of variability.

Results from this study suggest that using exotic parents that are significantly better in fiber elongation could improve the genetic base of adapted cultivars along with improving fiber elongation. Unlike other fiber properties, for fiber elongation an increase in exotic parentage percentage showed an almost linear response. Fiber elongation increased almost linearly with increase in exotic parentage percentage when the exotic parent had a significantly higher percent fiber elongation, and decreased almost linearly when the exotic parent had a significantly lower percent fiber elongation.

3.3.6. Short fiber content:

Tests for fixed effects revealed that population, exotic percentage, and interaction population × exotic percentage had a highly significant effect on short fiber content (Table 33). Covariance parameter estimates indicate that the nested effect (replication within environment), and interaction effect (population × replication within environment) were significant. Further breakdown of analysis reveal that the variation due to location played a significant role in replication within environment interaction. Significant environment, and or genotype × environmental interaction were reported in earlier studies (Jones et al., 2014; Wu et al., 2010; Zeng et al., 2010, 2011; Zeng and Meredith, Jr., 2009b). Increasing the number of testing locations may be required to adequately estimate short fiber content. Interactions (exotic percentage × replication within environment), and (population × exotic percentage × replication within environment) were not significant.

ANOVA results indicate population × exotic percentage interaction was significant, so means were compared among exotic parentage percentage groups (EP1, EP2, EP3 and EP4) and their respective adapted parents (EP0) within each population (Table 34). An increase in exotic parentage percentage had no effect on short fiber content in populations POP1, POP4 and POP6. In populations POP5 and POP8, an increase in exotic parentage percentage up to 25 percent reduced short fiber content significantly, but further increase in exotic content did not show any significant difference in population POP5. In population POP8, an increase in exotic parentage percentage beyond 25 percent did not show any significant difference from the adapted control except at 75 percent exotic parentage percentage. In POP2, an increase in exotic parentage

percentage up to 25 percent did not show a significant difference, but any further increase led to a significant increase in short fiber content. In populations POP3 and POP7, an increase in exotic percentage increased short fiber content at all levels except at 50 percent exotic parentage percentage in POP3, and at 75 percent exotic parentage percentage in PG7. These results agree with earlier studies where introgressed lines were not significantly different from cultivar checks (Zeng and Meredith, Jr., 2009a, 2009b).

Graphical illustration of short fiber content shows a clear difference between populations derived from exotic parents TX245 and TX1419 (Figure 18). Short fiber content values ranged from 7.5 to 9.5 percent among the populations derived from the exotic parent TX245, and 7.4 to 8.4 percent among populations derived from the exotic parent TX1419. Exotic parent TX245 performed inconsistently across experiments, whereas the exotic parent TX1419 performed consistently across experiments. A differential response by these two exotic parents across experiments indicates that exotic parent TX245 was more affected by environmental interaction than exotic parent TX1419.

Results from this study suggest that the effect of exotic introgression on short fiber content is difficult to predict. For the most part these results indicate that the genetic base of adapted parents can be improved without affecting short fiber content. Although exotic parents were not significantly different from adapted parents in 6 out of 8 populations, appearance of some lines with significantly increased short fiber content percentages indicate that using exotic parents with similar short fiber content as of adapted parents may not necessarily result in reducing short fiber content. However, there is higher chance of recovering lines which are not significantly different from adapted parents in short fiber content, which is evident from these results.

Table 23. ANOVA results and covariance parameter estimates for fiber length

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	44	151.20**	≤0.0001
Exotic Percentage	4	28	173.76**	≤0.0001
Population × Exotic Percentage	28	169	25.59**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.025*	0.014	1.802	0.0403
Pop×Rep(Environment)	0.001	0.001	1.716	0.0883
ExoPct×Rep(Environment)	0.001	0.001	1.291	0.1214
Pop×ExoPct×Rep(Environment)	-0.001	0.001		

*Significant at P ≤0.05

** Significant at P ≤0.01

Table 24. Least square mean comparisons for fiber length

	Fiber length (mm)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**28.45**	**25.91**	**29.21**	**27.43**	**28.45**	**26.16**	**29.46**	**27.43**
EP1	-2.29**	-1.52**	-4.32**	-1.78**	0.25	0.00	-1.78**	0.25
EP2	-3.30**	-2.79**	-4.32**	-3.81**	-2.03**	1.27**	-2.29**	-0.51
EP3	-4.83**	-3.05**	-5.84**	-4.06**	-1.78**	-0.25	-1.52**	0.00
EP4	-4.06**	-1.78**	-5.84**	-3.81**	-2.03**	0.00	-3.81**	-1.27**
SE†	0.25	0.25	0.25	0.25	0.25	0.25	0.25	0.25

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

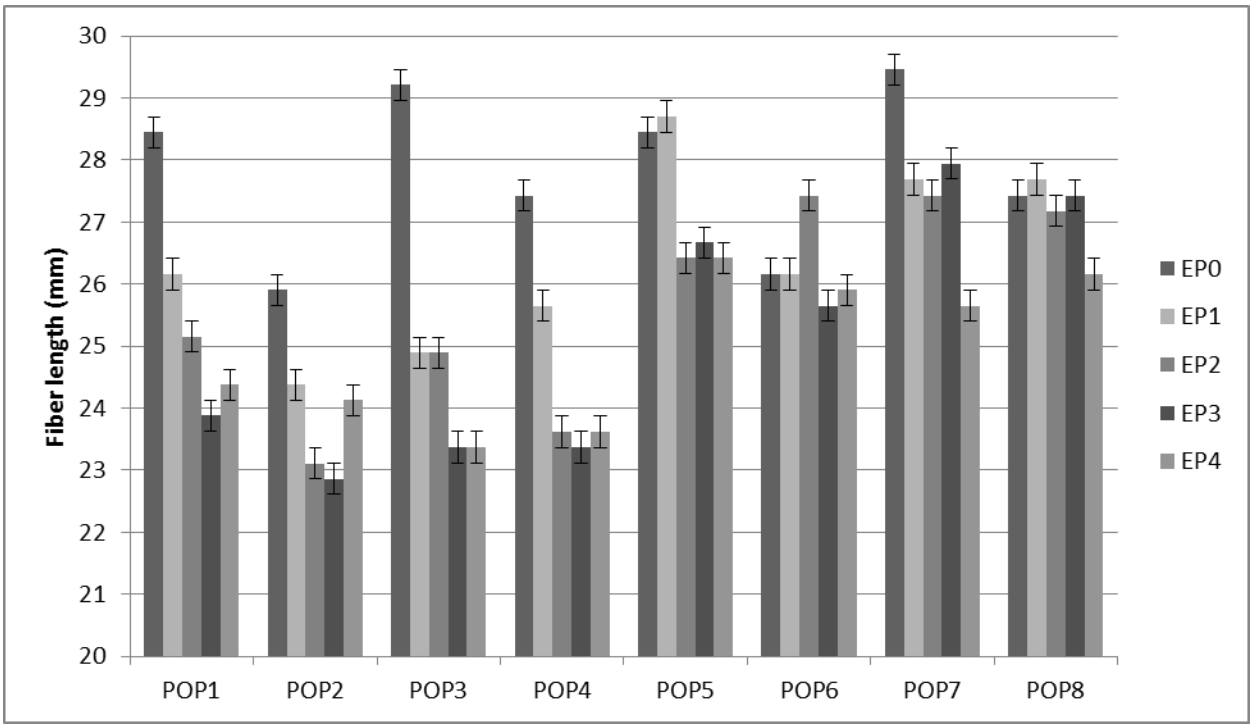

Figure 13. Effect of exotic germplasm introgression on fiber length. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 25. ANOVA results and covariance parameter estimates for fiber strength

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	44	41.57**	≤0.0001
Exotic Percentage	4	28	28.19**	≤0.0001
Population × Exotic Percentage	28	169	19.13**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.002	0.042	0.047	
Pop×Rep(Environment)	0.200*	0.081	2.475	0.0298
ExoPct×Rep(Environment)	0.064	0.049	1.303	0.0906
Pop×ExoPct×Rep(Environment)	0.035	0.083	0.419	

*Significant at P ≤.05

**Significant at P ≤0.01

Table 26. Least square mean comparisons for fiber strength

	Fiber strength (g tex^{-1})							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	32.83	27.94	31.97	27.34	32.01	27.95	31.94	28.30
EP1	-3.52**	-1.37**	-4.10**	0.01	0.13	-0.60	-3.15**	1.17**
EP2	-4.54**	-1.73**	-4.25**	-1.24**	-2.71**	2.29**	-2.49**	1.13**
EP3	-5.76**	-0.90*	-5.06**	-1.57**	-2.47**	0.40	-0.92**	2.67**
EP4	-5.44**	-0.62	-4.29**	0.11	-1.96**	2.33**	-1.65**	1.46**
SE†	0.50	0.36	0.39	0.37	0.53	0.44	0.43	0.44

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

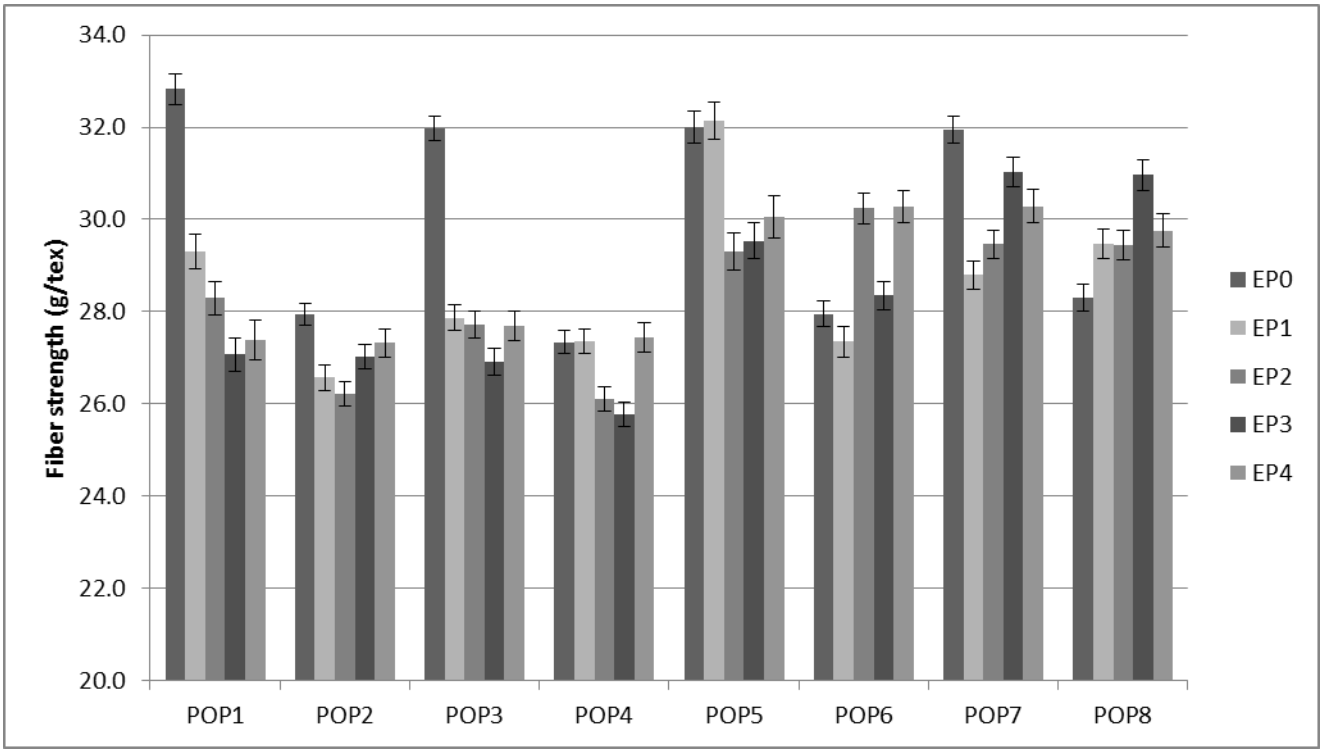

Figure 14. Effect of exotic germplasm introgression on fiber strength. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 27. ANOVA results and covariance parameter estimates for micronaire

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	44	8.78**	≤0.0001
Exotic Percentage	4	28	76.04**	≤0.0001
Population × Exotic Percentage	28	169	4.80**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.157*	0.086	1.826	0.0377
Pop×Rep(Environment)	0.016**	0.005	3.272	0.0063
ExoPct×Rep(Environment)	0.001	0.002	0.869	0.1507
Pop×ExoPct×Rep(Environment)	0.002	0.004	0.653	0.0556

*Significant at P ≤0.05
**Significant at P ≤0.01

Table 28. Least square mean comparisons for micronaire

	micronaire							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	4.53	5.07	4.61	4.78	4.52	4.94	4.50	4.87
EP1	0.28**	0.26*	0.70**	0.32*	0.18	-0.12	0.24	-0.13
EP2	0.54**	0.35*	0.58**	0.44**	0.48**	0.07	0.45**	-0.07
EP3	0.76**	0.34*	0.92**	0.60**	0.40**	0.20	0.59**	0.28*
EP4	0.73**	0.41**	0.97**	0.75**	0.73**	0.37*	0.97**	0.46**
SE†	0.10	0.12	0.10	0.14	0.10	0.15	0.12	0.14

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

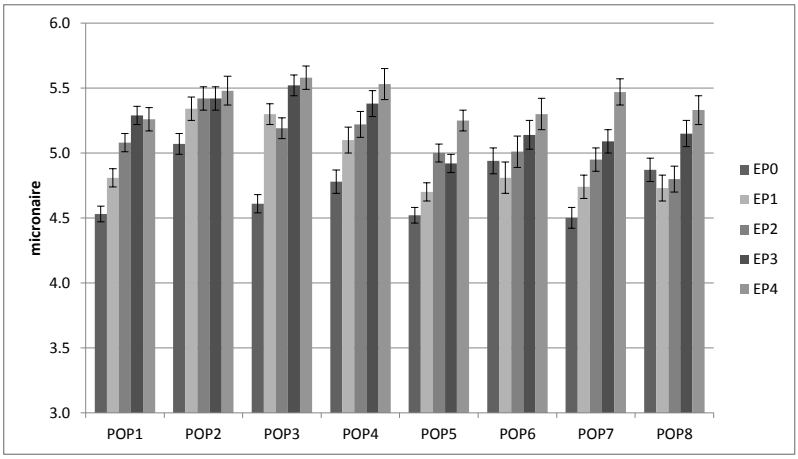

Figure 15. Effect of exotic germplasm introgression on micronaire. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 29. ANOVA results and covariance parameter estimates for fiber uniformity

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	44	12.23**	≤0.0001
Exotic Percentage	4	28	32.23**	≤0.0001
Population × Exotic Percentage	28	169	4.03**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.856*	0.475	1.802	0.0425
Pop×Rep(Environment)	0.094*	0.033	2.846	0.0147
ExoPct×Rep(Environment)	0.038	0.021	1.819	0.0531
Pop×ExoPct×Rep(Environment)	-0.005	0.030		

*Significant at P ≤0.05
**Significant at P ≤0.01

Table 30. Least square mean comparisons for fiber uniformity

	Fiber uniformity (%)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	**83.56**	**82.47**	**83.12**	**82.34**	**83.40**	**82.73**	**83.37**	**82.70**
EP1	-0.74	-0.93*	-1.52**	0.14	0.09	0.24	-1.13**	0.43
EP2	-1.18**	-1.56**	-1.18**	-0.84*	-1.11**	-0.16	-1.34**	-0.19
EP3	-2.31**	-1.80**	-1.95**	-0.80*	-1.40**	-0.59	-1.21**	-0.31
EP4	-2.02**	-1.31**	-2.45**	-0.99*	-1.30**	-0.67*	-1.39**	-0.48
SE†	0.39	0.37	0.35	0.37	0.29	0.32	0.29	0.28

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

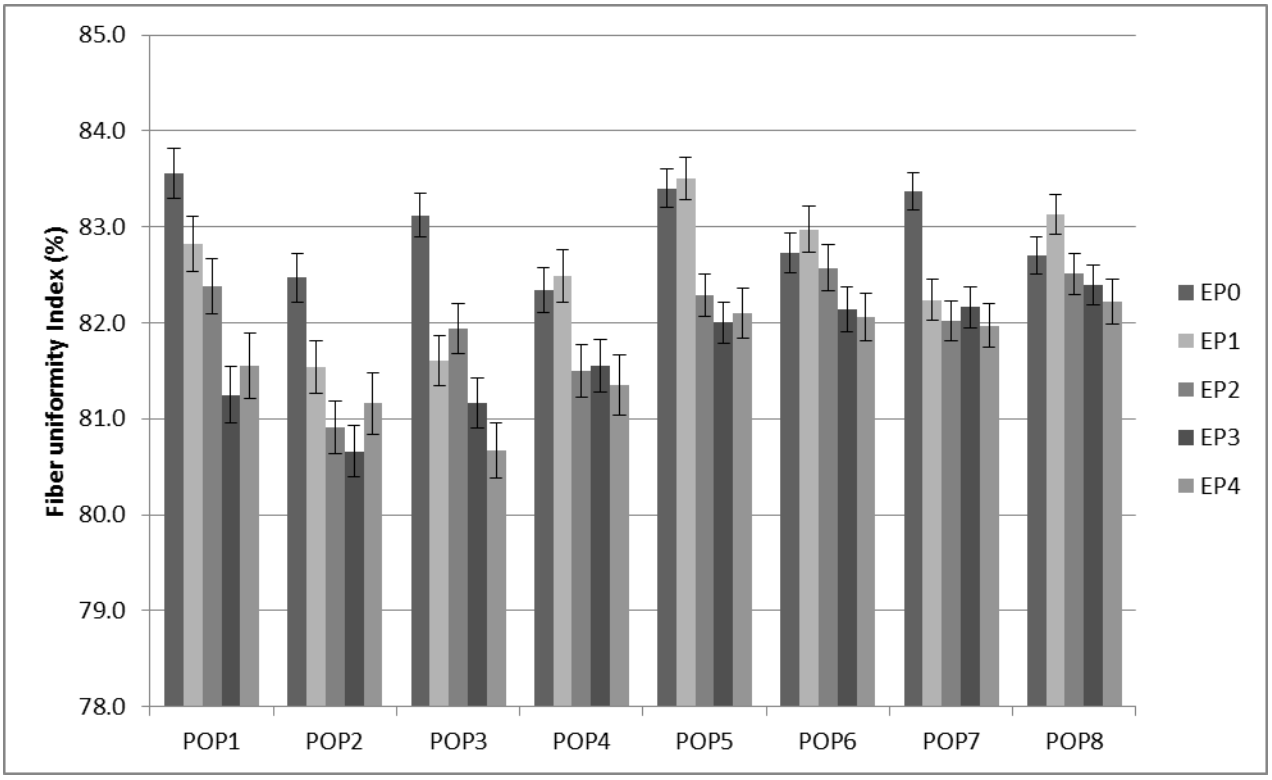

Figure 16. Effect of exotic germplasm introgression on fiber uniformity. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

Table 31. ANOVA results and covariance parameter estimates for fiber elongation

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	44	25.27**	≤0.0001
Exotic Percentage	4	28	89.87**	≤0.0001
Population × Exotic Percentage	28	169	18.60**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.142	0.081	1.763	0.0506
Pop×Rep(Environment)	0.047*	0.014	3.364	0.0101
ExoPct×Rep(Environment)	0.001	0.004	0.368	0.3564
Pop×ExoPct×Rep(Environment)	-0.009	0.010		

*Significant at P ≤0.05
**Significant at P ≤0.01

Table 32. Least square mean comparisons for fiber elongation

	Fiber elongation (%)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	3.93	5.56	4.33	6.56	4.08	5.89	4.39	6.29
EP1	1.36**	0.44*	1.28**	-0.3	0.59**	0.03	1.23**	-0.46**
EP2	1.23**	0.91**	1.55**	-0.06	1.07**	0.17	0.85**	-0.41**
EP3	1.77**	0.92**	1.77**	0.27	1.35**	-0.45**	0.90**	-0.51**
EP4	2.59**	1.01**	2.15**	-0.09	1.91**	-0.43**	1.53**	-0.65**
SE†	0.23	0.20	0.15	0.23	0.16	0.17	0.17	0.16

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

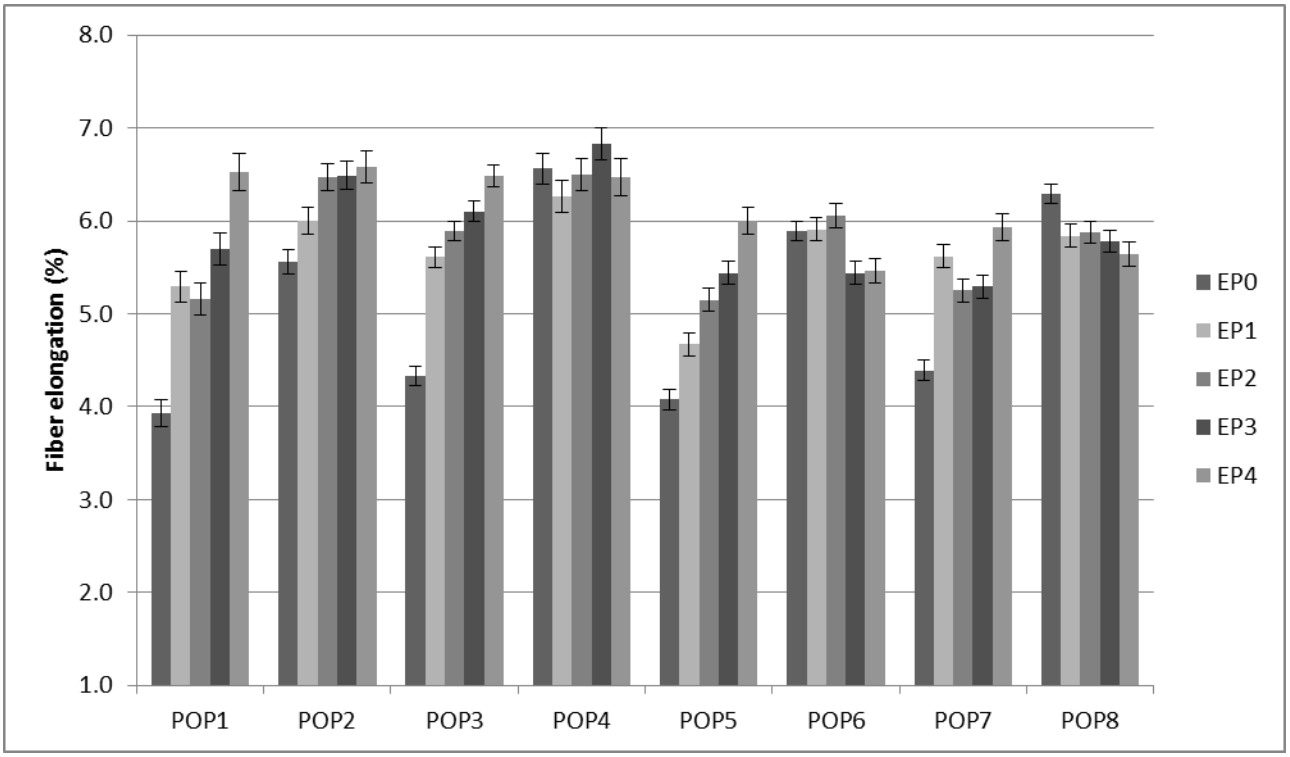

Figure 17. Effect of exotic germplasm introgression on fiber elongation. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic).

Table 33. ANOVA results and covariance parameter estimates for short fiber content

Fixed effects				
Source	**Num DF**	**Den DF**	**F value**	**P value**
Population	7	44	10.88**	≤0.0001
Exotic Percentage	4	28	3.90*	0.0123
Population × Exotic Percentage	28	169	3.87**	≤0.0001
Covariance parameter estimates (Random effects)				
Source	**Estimate**	**SE**	**Z value**	**P value**
Rep(Environment)	0.562*	0.313	1.795	0.0446
Pop×Rep(Environment)	0.068*	0.025	2.785	0.0146
ExoPct×Rep(Environment)	0.029	0.016	1.800	0.0533
Pop×ExoPct×Rep(Environment)	0.016	0.024	0.665	

*Significant at P ≤0.05
**Significant at P ≤0.01

Table 34. Least square mean comparisons for short fiber content

	Short fiber content (%)							
	POP1	**POP2**	**POP3**	**POP4**	**POP5**	**POP6**	**POP7**	**POP8**
EP0	7.67	8.24	8.09	8.90	7.98	8.01	7.77	8.35
EP1	-0.06	0.59	0.67*	-0.54	-0.57*	-0.14	0.55*	-0.41*
EP2	0.15	1.10**	0.35	0.41	0.07	-0.33	0.68*	-0.35
EP3	0.61	0.84*	0.98**	-0.07	0.41	0.22	0.32	-0.56*
EP4	0.46	0.07	1.39**	-0.26	0.02	0.07	0.62*	-0.34
SE†	0.31	0.36	0.31	0.32	0.21	0.24	0.27	0.19

ǂ EP0 = 0 %, EP1 = 25%, EP2 = 50%, EP3 = 75%, EP4 = 100% exotic germplasm.

† Standard error

* Significant at P ≤0.05

** Significant at P ≤0.01

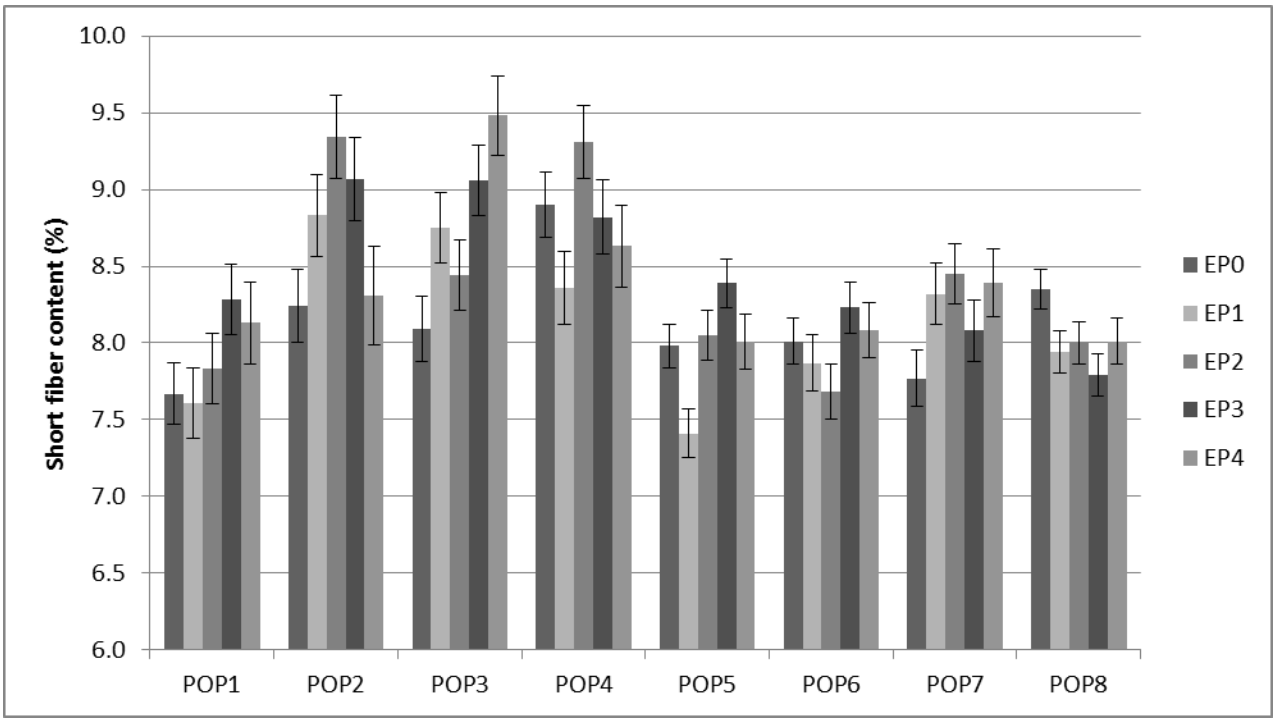

Figure 18. Effect of exotic germplasm introgression on short fiber content. Populations (POP 1-8), Exotic percentage levels (EP0 (0% exotic), EP1 (25% exotic), EP2 (50% exotic), EP3 (75% exotic) and EP4 (100% exotic)).

CONCLUSIONS

An increase in exotic germplasm percentage significantly lowered agronomic performance, however the magnitude of the effect varied with populations and exotic percentage levels. Among agronomic traits seed cotton yield, lint seed^{-1}, lint percentage and lint yields were the most affected, whereas days to first flower, bolls plant^{-1} and lodging were traits least affected with an increase in exotic percentage. Boll size and seeds boll^{-1} were unaffected or improved in populations (except in POP1) derived from exotic parent TX245, whereas both traits declined significantly in populations derived from exotic parent TX1419. In most populations an increase in exotic parentage percentage did not show any significant difference from the adapted parent up to 25 percent exotic germplasm introgression for all the agronomic traits except for lint seed^{-1}. In almost all the cases where exotic the parent was significantly better, no significant difference was observed up to 25 percent suggesting that an increase in exotic germplasm can expand genetic base without adversely affecting agronomic traits.

An increase in exotic germplasm percentage significantly lowered fiber properties, however the magnitude of the effect varied with populations and exotic percentage levels. Among fiber properties micronaire was the most affected trait, whereas fiber elongation and short fiber content were the least affected traits with an increase in exotic germplasm introgression. Populations derived from exotic parent TX245 showed a significant reduction in fiber length, fiber strength, micronaire, and fiber uniformity, but fiber elongation improved with an increase in exotic parentage percentage and remained unaffected in most cases for short fiber content. Populations derived from exotic parent TX1419 gave mixed results with an increase in

exotic parentage percentage. Except for fiber elongation, fiber properties remained unaffected or improved in POP6 and POP8 with an increase in exotic parentage percentage. Fiber strength improved significantly with an increase in exotic germplasm in POP8. In almost all cases where the exotic parent was better or equal with performance of adapted cultivars, an increase in exotic parentage percentage up to 25 percent did not show a significant difference in fiber properties, which suggests that using exotic parents which are similar or better in fiber properties could expand genetic base of adapted cultivars without altering fiber properties.

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