

**SELECTION OF EARLY GENOTYPES BY BIOMETRICAL AND MOLECULAR
TOOLS TO IMPROVE FIBER YIELD IN UPLAND COTTON (*Gossypium hirsutum* L.)**

A PhD DISSERTATION

BY

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Registration No: 1505198

DOCTOR OF PHILOSOPHY

IN

GENETICS AND PLANT BREEDING



DEPARTMENT OF GENETICS AND PLANT BREEDING

HAJEE MOHAMMAD DANESH SCIENCE AND TECHNOLOGY UNIVERSITY

DINAJPUR-5200, BANGLADESH

DECEMBER 2025

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Certification

This is to certify that the dissertation entitled “**SELECTION OF EARLY GENOTYPES BY BIOMETRICAL AND MOLECULAR TOOLS TO IMPROVE FIBER YIELD IN UPLAND COTTON (*Gossypium hirsutum* L.)**” a study, prepared by the examinee, bearing the Registration No.: 1505198, Department of Genetics and Plant Breeding, Hajee Mohammad Danesh Science and Technology University, Dinajpur for the award the degree, Doctor of Philosophy in Genetics and Plant Breeding, is a record of original research work carried out by the author under my supervision. The work is an original and unique one and to the best of my knowledge, any part of the dissertation has yet not been produced elsewhere for any degree.

(Professor Dr. Md. Arifuzzaman)

Supervisor

DECLARATION

I do hereby declare that the dissertation entitled **“SELECTION OF EARLY GENOTYPES BY BIOMETRICAL AND MOLECULAR TOOLS TO IMPROVE FIBER YIELD IN UPLAND COTTON (*Gossypium hirsutum* L.)”** submitted to the Hajee Mohammad Danesh Science and Technology University (HSTU), Dinajpur, Bangladesh for the Degree of Doctor of Philosophy was entirely own research work, outlined and carried out by me and original. The dissertation or its part in any form has neither been submitted anywhere else for obtaining any degree nor does it contain results of any collaborative study except my research work.

The Author

HSTU, Dinajpur

December 2025

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The Author

*This humble effort is
dedicated to
My Beloved Daughter
Rumaísa Tasnīm
and
My Parents*

SELECTION OF EARLY GENOTYPES BY BIOMETRICAL AND MOLECULAR TOOLS TO IMPROVE FIBER YIELD IN UPLAND COTTON (*Gossypium hirsutum* L.)

ABSTRACT

Cotton (*Gossypium hirsutum* L.) is a major fiber crop but contributing minimum to our GDP. However, productivity remains limited due to long crop duration (6-7 months) and low yield of cultivated varieties. Therefore, this study aimed to identify early-boll picking with high quality fiber yield potential genotypes through integrated biometrical and molecular approaches. Three experiments were conducted from July 2021 to December 2023 at Cotton Research, Training and Seed Multiplication Farm, Sadarpur, Dinajpur started with 100 and ended to 12 upland cotton genotypes comprising released varieties, elite breeding lines, local cultivars, a mutant variety, and advanced lines. In the first experiment, genotypes JA-11/L, BC-0025, BC-0033, BC-0027, RA-2, and BC-0024 showed superior yield and early performance. High heritability coupled with high genetic advance was shown by boll number, boll weight, and seed cotton yield. Correlation and path analyses identified sympodial branches, bolls per plant, and single boll weight as key yield determinants. Cluster and principal component analyses grouped the genotypes into eight distinct clusters, with the first five principal components explaining 73.09% of total variation. The second experiment evaluated 45 selected genotypes for earliness and productivity. Genotypes RA-2, BC-0027, BC-0491, BC-0510, and BC-0512 combined early maturity with high yield and outperformed standard checks. The third experiment assessed 12 promising genotypes for morphological, fiber quality traits and molecular profiling. Significant variation was detected in plant height, flowering time, bolls per plant, fiber yield, and earliness index. Fiber quality parameters like upper half mean length, strength, micronaire, and reflectance varied significantly. The genotype BC-0491 and BC-0510 exhibited superior fiber attributes. Molecular characterization using 20 SSR markers identified nine polymorphic markers generating 26 alleles. Markers MGHE16 and NAU3736 showed high polymorphic information content. STRUCTURE and UPGMA analyses revealed two major genetic clusters, confirming genetic distinctness among the selected genotypes. Overall, integration of morphological, fiber, and molecular analyses identified the genotypes RA-2, BC-0027, BC-0491, BC-0510, and BC-0512 as promising parental lines for developing early-maturing, high-yielding cotton varieties with improved fiber quality suitable for Bangladesh.

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ACRONYMS AND ABBREVIATION

Abbreviations/symbols	Meanings
ADE	Additive-dominance-environment
AEZ	Agro Ecological Zone
AFLP	Amplified fragment length polymorphism
ANOVA	Analysis of variance
BBS	Bangladesh Bureau of Statistics
CD	Critical difference
CDB	Cotton Development Board
CG	Cotton grade
Cm	Centimeter
CMD	Cotton marker database
CV	Coefficient of variation
DAP	Days after planting
d.f.	Degrees of freedom
DNA	Deoxyribonucleic acid
EDTA	Ethylene diamine tetra acetic acid
<i>et. al.</i>	<i>Et alu</i> = Other people
FAO	Food and Agricultural Organization
FAS	Foreign Agriculture Service
FST	Fixation index
GOT	Ginning out turn
HSTU	Hajee Mohammad Danesh Science and Technology University
HVI	High Volume Instrument
Ha	Hactare
h^2_b	Heritability in broad sense
h^2_n	Heritability in narrow sense
g	Gram
GA	Genetic advance
GAM	Genetic advance as percentage of mean
GCV	Genotypic coefficient of variation
ICAC	International Cotton Advisory Committee
i.e.	id est (That is)
IPM	Integrated pest management
IRRI	International Rice Research Institute
Kg	Kilogram
L	Liter
LSD	Least significant difference
Mb	Megabase
Mic.	Micronaire
ML	Mean length
mm	Milimeter
mM	Milimolar
MS	Mean Sum of Square

ACRONYMS AND ABBREVIATION

Abbreviations/symbols	Meanings
MS _E	Error mean square
MY	Marketing Year
MYT	Multilocation yield trial
PC	Principal component
PCA	Principal component analysis
PCR	Polymerase chain reaction
PCV	Phenotypic coefficient of variation
PIC	Polymorphic information content
pH	Negative logarithm of hydrogen ion (H ⁺) concentration
QTL	Quantitative Trait Loci
RAPD	Random amplification of polymorphic DNA
Rd	Reflectance
RFLP	Restriction fragment length polymorphism
RCBD	Randomized Complete Block Design
RH	Relative Humidity
R&D	Research and Development
r _p	Phenotypic correlation
r _g	Genotypic correlation
SE	Standard error
SFI	Short fiber index
SSR	Simple sequence repeat
SNP	Single nucleotide polymorphism
Str.	Strength
TBE	Tris-borate-EDTA
Tex	Texture
TSP	Triple super phosphate
UHML	Upper half mean length
UI	Uniformity index
UPGMA	Unweighted pair group method with arithmetic mean
USD	United States dollar
USDA	United States Department of Agriculture
Viz.	Namely
WIA	Wilmotts's index of agreement
μg	Microgram
μL	Microliter
+b	Yellowness
°C	Degree centigrade
σ ² _A	Additive variance
σ ² _D	Dominance variance
σ ² _e	Error variance
σ ² _g	Genotypic variance
σ ² _p	Phenotypic variance

CHAPTER I

INTRODUCTION

Cotton (*Gossypium* spp.) is a globally important fiber crop with substantial economic and industrial value. Archaeological evidence indicates its domestication 5,000–6,000 years ago in the Indus Valley, Egypt, and Central America (Smith & Stephens, 2011; Brubaker *et al.*, 1999). Today, it is widely cultivated in tropical and subtropical regions due to its broad agro-ecological adaptability. Beyond textile fiber, cotton provides edible oil and protein-rich seed cake, supporting millions of smallholder farmers and contributing significantly to national economies (Kadam *et al.*, 2024; Kranthi, 2018; Dinakaran *et al.*, 2012).

The genus *Gossypium* (Malvaceae) comprises 53 recognized species, including 46 diploids ($2n = 2x = 26$) and 7 allotetraploids ($2n = 4x = 52$; AADD) (Kranthi, 2018; Chen *et al.*, 2007; Ulloa, 2006). The principal cultivated species are the tetraploids *G. hirsutum* and *G. barbadense*, while *G. arboreum* and *G. herbaceum* are important diploids. Diploid genomes are ~880 Mb (Hawkins *et al.*, 2006; Hendrix & Stewart, 2005), whereas tetraploid genomes are larger (~2,300 Mb), reflecting ancient polyploidization events (Desai *et al.*, 2006; Wendel, 1989). The genus *Gossypium* is unique due to the independent domestication of four species for their seed fiber trichomes: two Old World species (*G. arboreum* and *G. herbaceum*) originating from Africa, Egypt, and the Indian subcontinent, and two New World species (*G. hirsutum* and *G. barbadense*) from the Americas (Wendel & Grover, 2015; Smith & Stephens, 2011; Wendel & Cronn, 2003). Polyploidy in cotton not only expanded the genetic repertoire but also enhanced agronomic traits such as fiber length, strength, and yield potential, which are key factors for the success of tetraploid species in global cultivation.

Cotton (*Gossypium hirsutum* L.) is the most widely cultivated fiber crop, grown in over 80 countries and contributing more than 95% of global cotton production (Farias *et al.*, 2016; Shakeel *et al.*, 2015). Its broad adaptability, high yield potential, and desirable fiber attributes underpin its dominance, whereas *G. barbadense* is valued for premium extra-long staple fiber. Beyond textile use, cotton is a multifunctional crop: ginning separates lint for yarn production (Cotton.org, 2025), while seeds provide edible oil and

protein-rich meal, and linters ($\approx 5\text{--}6\%$ of seed weight) serve as raw materials for paper, cellulose derivatives, biodegradable plastics, and pharmaceutical applications (Wiley Online Library, 2009; Salama *et al.*, 2024; Li *et al.*, 2015). Globally, cotton supports over 60 million livelihoods and sustains a textile economy valued at nearly USD 3 trillion (Dinakaran *et al.*, 2012; Kadam *et al.*, 2024).

In Bangladesh, upland cotton accounts for approximately 96% of national production (Tabib, 2023); however, average lint yield ($1,090 \text{ kg ha}^{-1}$ in 2020–21) remains considerably lower than that of China ($1,787 \text{ kg ha}^{-1}$), Australia ($1,887 \text{ kg ha}^{-1}$), and Brazil ($1,712 \text{ kg ha}^{-1}$) (ICAC, 2022). This yield gap is largely attributed to a narrow genetic base, limited adoption of improved cultivars, and suboptimal agronomic practices (Malik *et al.*, 2014; Nazir *et al.*, 2013). Since cotton yield and fiber quality traits—such as staple length, strength, fineness, maturity, and uniformity—are quantitatively inherited and environmentally influenced (Ali *et al.*, 2008; Balçı *et al.*, 2020), effective improvement depends on systematic assessment of genetic variability.

Estimation of genetic parameters, including genotypic and phenotypic coefficients of variation, heritability, and genetic advance, provides insight into trait inheritance and selection efficiency (Batoool *et al.*, 2010; Dhamayanathi *et al.*, 2010; Khan *et al.*, 2010). Significant variability coupled with high heritability and genetic advance has been reported for yield-contributing traits in cotton (Abbas *et al.*, 2015; Ahsan *et al.*, 2015). Correlation and path coefficient analyses further partition direct and indirect effects of component traits on yield, enabling precise trait prioritization in breeding programs (Wright, 1921; Dewey & Lu, 1959; Sainath *et al.*, 2022; Kadam *et al.*, 2024; Sharma *et al.*, 2023; Manonmani *et al.*, 2019).

Breeding programs seek to elucidate the interrelationships among morphological traits to enhance selection efficiency (Khan *et al.*, 2017; Raza *et al.*, 2016; Reddy *et al.*, 2016). A comprehensive understanding of the nature and magnitude of trait associations with yield is essential for identifying superior parents for hybridization (Abdel-Monaem *et al.*, 2022; Teklewold *et al.*, 2000), particularly by emphasizing traits with positive contributions to yield improvement (Huang *et al.*, 2017; Sun *et al.*, 2017).

Genetic diversity, essential for long-term genetic gain and stress resilience (Salgotra & Chauhan, 2023; Swarup *et al.*, 2021), has been constrained in cultivated cotton due to

domestication bottlenecks and repeated selection (Liang *et al.*, 2017; Hinze *et al.*, 2015). Multivariate approaches such as hierarchical clustering particularly Ward's method (Ward, 1963) and principal component analysis (PCA) are widely used to quantify phenotypic divergence and population structure (Brown-Guedira *et al.*, 2000; Jarwar *et al.*, 2019b; Kakar *et al.*, 2021; Jaslam *et al.*, 2022; Sudeepthi *et al.*, 2018). These methods have been effectively applied in cotton diversity studies (Nadeem *et al.*, 2022; Sun *et al.*, 2019; Shabbir *et al.*, 2016; Shakeel *et al.*, 2015; Nazir *et al.*, 2013).

However, morphological traits alone provide limited resolution. Molecular markers, particularly simple sequence repeats (SSRs), offer co-dominant inheritance, high polymorphism, reproducibility, and genome-wide distribution, making them powerful tools for genetic diversity analysis and breeding applications (Rakshit *et al.*, 2010; Ali *et al.*, 2011; Ahmed *et al.*, 2013). SSRs have been extensively utilized in cotton for diversity assessment and QTL mapping (Kalivas *et al.*, 2011; Kuang *et al.*, 2011; Liang *et al.*, 2012; Li *et al.*, 2014). Currently, over 17,448 SSR markers are cataloged in cotton marker databases (CMD, <http://www.cottonmarker.org>).

Given the persistent yield stagnation and narrow genetic base of cotton in Bangladesh, integrated evaluation combining morpho-agronomic traits, multivariate statistical analyses, and molecular characterization is imperative. Comprehensive germplasm assessment will accelerate the identification of genetically diverse, high-yielding, and early-maturing genotypes suitable for sustainable cotton production in Bangladesh and comparable agro-ecological regions.

Based on the above rationale, the objectives of the present study were-

1. To select superior genotypes based on early maturing, yield and fiber quality traits.
2. To evaluate genetic diversity among the cotton genotypes.
3. To assess molecular characterization and phylogenetic relationships using SSR markers.
4. To emphasize the potential applicability of early boll picking and high seed cotton yielding with superior fiber quality cotton genotypes in national cotton improvement programs.

CHAPTER II

REVIEW OF LITERATURE

Gossypium hirsutum L., commonly known as upland cotton or American cotton, is the most widely cultivated cotton species globally, accounting for approximately 90% of total world cotton production and about 95% of production in the United States, the largest global exporter (ICAC's 82nd Plenary Meeting, 2024; FAO, 2023; ICAC, 2022; Smith & Stephens, 2011). Belonging to the family Malvaceae, it is a tetraploid species ($2n = 4x = 52$) widely cultivated for its adaptability, yield potential, and fibre quality.

2.1. Origin and evolution of cotton

The genus *Gossypium* comprises over 53 species distributed in tropical and subtropical regions. Among these, four are cultivated: *G. arboreum* and *G. herbaceum* (Old World diploids, $2n = 26$), and *G. hirsutum* and *G. barbadense* (New World allotetraploids, $2n = 52$) (Wendel *et al.*, 1992; Wendel, 1989).

G. hirsutum originated through allopolyploidization between an A-genome ancestor (African/Asian origin) and a D-genome ancestor (American origin) approximately 1–2 million years ago (Huang *et al.*, 2020). This hybridization likely occurred in the Yucatán Peninsula or southern Mexico, forming the AD genome group (Grover *et al.*, 2012; Li *et al.*, 2015). Wild progenitors of *G. hirsutum* are distributed across southern Mexico, Guatemala, and coastal Central America, where high genetic diversity persists in several wild races such as *yucatanense*, *latifolium*, and *marie-galante* (Wegier *et al.*, 2011).

2.2. Domestication, diversification and global distribution of cotton

Archaeobotanical and genomic evidence indicate that *G. hirsutum* was domesticated in southern Mexico approximately 4,000–5,000 years ago (De Montellano, 2012; Brubaker *et al.*, 1999). Domestication involved selection for lint length, fibre strength, synchronous maturity, and photoperiod insensitivity.

Following its domestication, *G. hirsutum* diversified into numerous cultivated forms and landraces, which later spread to the Caribbean Islands and South America, becoming the basis for most modern cultivars (Wendel & Cronn, 2003). *G. hirsutum* is now cultivated throughout tropical and subtropical regions of the world. Major producing countries

include China, India, the United States, Pakistan, Brazil, Uzbekistan, Turkey, and Australia (FAO, 2023). It thrives under both irrigated and rainfed conditions, demonstrating exceptional environmental plasticity. Domesticated thousands of years ago, it has since become the world's predominant cotton species, cultivated across nearly all cotton-growing regions due to its superior adaptability and productivity. Owing to its adaptability and fibre versatility, *G. hirsutum* forms the genetic backbone of modern cotton breeding programmes.

2.3. Taxonomy of upland cotton

Gossypium hirsutum L., commonly known as upland or American cotton exhibits extensive morphological and genetic variability, encompassing numerous varieties and hybrid cultivars with differences in fiber length, environmental adaptability, and yield potential; long-staple upland types are particularly valued for superior fiber quality and commercial use (Fryxell, 1979). In addition to its primary role as a fiber crop, *G. hirsutum* along with *G. herbaceum* serves as a key source of cottonseed oil (FAO, 2023). Ethnobotanical evidence also highlights its cultural significance, such as its traditional use by the Zuni people for ceremonial garments and cordage (Castetter & Underhill, 1935). Biologically, the species is characterized by the presence of extrafloral nectaries, which play ecological roles in plant–insect interactions (Rudgers & Gardener, 2004).

Scientific Name: *Gossypium hirsutum* L. (Kartesz, 1994)

Other Common Names: Cotton (EN), upland cotton (EN)

Kingdom: Plantae (Plants)

Phylum: Anthophyta (Flowering plants)

Class: Dicotyledoneae (Dicotyledons/Eudicots)

Order: Malvales

Family: Malvaceae (Mallow family- known for fibrous, mucilaginous plants)

Subfamily: Malvoideae

Genus: *Gossypium* (Cotton plants- includes >50 species)

Species: *Gossypium hirsutum*

2.4. An overview of the evolution and contemporary status of cotton cultivation programs in Bangladesh

Bangladesh has a long heritage in textile production, historically exemplified by Bengal's world-famous Muslin fabric. Colonial disruptions during the British period dismantled indigenous weaving and raw-cotton systems by the early nineteenth century (Ahmed, 1995). After independence in 1971, the Cotton Development Board (CDB) was established to revive cotton through varietal trials, research, and extension programs (CDB, 2007) (**Figure 1**).

At present, cotton cultivation covers about 45,000 ha, producing 150,000–200,000 bales annually, only 3–4 % of national demand (Dristy *et al.*, 2024). In contrast, the RMG-driven textile industry consumes 7.5–8 million bales each year, largely imported from the United States, Brazil, and West Africa making Bangladesh one of the world's largest cotton importers (USDA FAS, 2024). Bangladesh's cotton consumption in MY 2024-25 was forecasted to rise to 7.8 million bales, with ~7.7 million of those were expected to be imported (Local media/trade press, 2025). Imports are projected to reach ~8.1 million bales by 2025–26, underscoring persistent structural dependence on external sources (USDA FAS, 2025; Biz BD Review, 2025).

Domestic expansion remains constrained by limited arable land, competition with food crops, low-yielding and long-duration varieties, pest incidence, and inadequate mechanization and irrigation (Dristy *et al.*, 2024). Current policies focus on short-duration, high-yield varieties, improved seed systems, and sustainable intensification to reduce import reliance under changing climatic conditions (Dristy *et al.*, 2024; Cotton Development Board, 2007).

2.5 Review of literature on analysis of variance and mean performances of different traits in upland cotton

Beegum *et al.* (2025) conducted a study to analyze fiber quality data from 40 upland cotton cultivars, including TM1. The measured fiber quality from all cultivars was then compared with the model simulated fiber quality. Results Among the 40 upland cultivars, fiber strength varied from 28.4 cN·tex⁻¹ to 34.6 cN·tex⁻¹, fiber length ranged from 27.1 mm to 33.3 mm, micronaire value ranged from 2.7 to 4.6, and length uniformity index varied from 82.3% to 85.5%. The model simulated fiber quality closely

COTTON PRODUCTION IN BANGLADESH

- CDB's unit offices representing cotton growing area
- ▲ CDB's zonal offices
- CDB's regional Office
- CDB's Research, Seed multiplication and Training Farm

Figure 1. The Map showing cotton growing areas in Bangladesh

matched the measured values for TM1, with the absolute percentage error (APE) being less than 0.92% for fiber strength, fiber length, and length uniformity index and 4.7% for micronaire. However, significant differences were observed for the other cultivars.

Baghyalakshmi *et al.* (2024) studied genetic improvement of fiber quality in tetraploid cotton. Fiber length, fiber strength, and fiber fineness are three main attributes that contributed to the quality of cotton fibers.

Çakmak *et al.* (2024) aimed to combine yield and fiber quality by broadening the genetic base of the genotypes. The mean and range of crosses were higher for seed cotton yield and ginning out-turn; similar for fiber strength and length; coarse for fiber fineness compared to their parents.

Makhmadjanov *et al.* (2024) studied the productivity of different cotton genotypes under conditions of Turkestan region of Kazakhstan to determine the most promising genotypes for commercial cultivation. Four genotypes were selected: Tashkent-6, Maktaral-5027, Namangan-1 and Bukhara-8. The study included analysis of such parameters as, plant height, flowering and maturity time, yield and number of bolls and fibre quality characteristics. Statistical analysis confirmed the significance of differences between genotypes. The results showed that Tashkent-6 and Maktaral-5027 had the best parameters: yield 3.8 t/ha and 3.5 t/ha, flowering time 55 days and maturity 120 days, plant height 120 cm and 115 cm, number of bolls 45 and 42. The fibre quality of these genotypes was also high: length 32 mm and 31 mm, tensile strength 30 g/tex and 29 g/tex, fineness 2.5 dtex and 2.6 dtex. Namangan-1 and Bukhara-8 genotypes showed moderate results: yield 3.2 t/ha and 3.0 t/ha, flowering time 60 days and ripening time 130 days, plant height 110 cm and 105 cm, number of bolls 40 and 38, fibre length 30 mm and 29 mm, tensile strength 28 g/tex and 27 g/tex, fineness 2.8 dtex and 2.9 dtex. The results of the study confirmed high productivity of genotypes Tashkent-6 and Maktaral-5027, which made them promising for commercial cultivation in Kazakhstan, while genotypes Namangan-1 and Bukhara-8 also had potential for certain agronomic conditions.

Vadodaria *et al.* (2024) conducted a study comprising of 30 *G. arboreum* accessions to estimate the genetic variability using morphological and fibre quality parameters. Presence of substantial genetic variability was revealed by analysis of variance (ANOVA) amongst all genotypes for all the traits. Morphological studies indicated that

genotype PA 255 had higher seed cotton yield (80.07 g/plant). Examination of fibre suggested that genotype Cernuum had the highest fibre fineness (7.27 μ g/inch) while genotypes 824 (29.57 mm) and PA 812 (29.30 g/tex) had the maximum UHML and fibre strength, respectively.

Amer & Hassan (2023) conducted an experiment at Sakha Station of Agricultural Research, over 2021 and 2022 seasons to evaluate the variance among some Egyptian cotton genotypes for yielding potential. The genotypes were arranged in RCBD design replicated quadric. Materials comprised thirteen extra-long staple cotton genotypes (*Gossypium barbadense* L.). Data were collected for, boll weight, seed cotton yield, lint yield, lint%, micronaire reading, fiber strength, fiber length earliness index and uniformity index. Genotypes (G) exhibited significant differences for all traits. Years (Y) had insignificant differences on most of traits, GxY interaction was significant for some traits. Giza 87 and Giza 88 had the lower yield while the rest of genotypes insignificantly varied. Giza 87, Giza 93 and Giza 93 x Menufi genotypes had the finest fibers, Giza 87, Giza 92 and Giza 96 had the strongest fibers.

Divya *et al.* (2023) studied thirty genotypes of cotton to estimate genetic variability for 15 traits during kharif, 2022. Analysis of variance showed significant amount of variation among the genotypes for traits studied.

Mahdy *et al.* (2023) conducted an experiment to study the gene action that controls yield and quality traits of Egyptian cotton under normal and saline soil. Nine lines were crossed to three testers in a line $\times$ tester mating design. The 27 crosses and their parents were evaluated under normal and saline soil conditions in the seasons of 2018 and 2019 using a randomized complete block design with three replicates. Under normal soil and based on the mean performance the cross (Giza90 $\times$ Aus $\times$ Giza85) $\times$ Giza95 was the best one for SCY/p, LY/p, NB/p, BW, SI and NS/b, and Ashmouni $\times$ Giza90 $\times$ Aus $\times$ Giza83 for DFF and lint%. Dandara $\times$ Giza90 $\times$ Aus $\times$ Giza83 for LI, Giza85 $\times$ Giza95 for UHML. Giza95, Giza90 and Giza90 $\times$ Aus $\times$ Giza83 were the best combiners in all the analyzed traits. These crosses might be used to develop new lines for the two environments.

Selvaraj *et al.* (2023) studied the relationship among the quality parameters. The observations were taken in the F₁ population of 35 hybrids along with lines (7 lines) and testers (5 testers) for the traits viz. ginning outturn (%), upper half mean length (cm),

elongation percent (%) and micronaire value (μg per inch). Path analysis study revealed that lint index (1.3526) and upper half mean length (0.3392) had high direct positive effects on single plant yield. Hence selection based on these traits that are positively associated and direct positive effect with the yield might produce the best outcome in the genotypes in further breeding programmes.

Subalakhshmi *et al.* (2023) investigated eight parents and 56 hybrids were evaluated for 18 economically significant traits. The study was conducted at Department of Cotton, Tamil Nadu Agricultural University, Coimbatore, in 2021-2022. Analysis of variance was highly significant for all the traits studied.

Vishnuvardhan *et al.* (2023) evaluated 17 cotton genotypes with a view to study of genetic parameters for different yield and yield parameters. The analysis of variance revealed the existence of significant differences among the traits studied.

Balarabe *et al.* (2022) evaluated one hundred cotton genotypes from diverse sources for genetic improvement of important yield and fibre quality traits in cotton. Hence, the need to study and explore available germplasm resources to assess the variation among the cotton germplasm and to classify and identify the potential genotypes that can be used in the development of hybrid with the best fibre quality traits. The experiment was conducted in a 10 x 10 lattice design, replicated twice across two environments in 2020 wet season. Significant ($p \leq 0.05$) genotypic variation was observed for plant height, number of sympodial branches, number of bolls, seed cotton yield and for important fibre quality traits like fibre length and fibre fineness. Using rank summation index, genotypes VIR-7112-HG-69-15, VIR 7072, LA-213-SEA-ISLAND LEAF, BULGARIA-996 and 30858 with rank summation indices of 243, 270, 313, 330, 332 and 368 respectively were the best materials and potential genotypes for the development of hybrid with the best agronomic and yield traits, while LINIA-7010 ranked last with rank summation index of 712.

Manan *et al.* (2022) investigated eight cotton genotypes with their 15 F_1 hybrids (five lines, three testers) which were grown in the field conditions under randomized complete block design (RCBD) with two treatments i.e. normal and high- temperature stresses with two replications followed by split-plot arrangement to understand the effect of heat stress on morphology and fiber quality of the cotton. Data were collected for yield-

related parameters and fiber quality parameters like lint%, seed index, short fiber index, fiber strength, fiber fineness and upper half mean length. Mean value of lint% increased under high-temperature. Mean values of all studied traits were reduced significantly under high-temperature stress.

Meena *et al.* (2022a) carried out an investigation using twenty-five genotypes for quantitative and qualitative traits of cotton to estimate the genetic variability present within genotypes. The material was evaluated in randomized block design with four replications during kharif, 2019 at Agronomy Instructional Farm, S. D. Agricultural University, Sardarkrushi nagar. The differences between genotypes were significant for all the characters, which revealed considerable amount of variability for all the traits under study.

Nadeem *et al.* (2022) investigated twenty exotic accessions of cotton to check their variability for within-boll yield components. The experiment was performed at the research area of the department of Plant Breeding and Genetics, University of Agriculture, Faisalabad. The genotypes were planted in three replications following a Randomized Complete Block Design (RCBD). At maturity, data were collected for different within-boll yield parameters. The analysis of variance showed significant results for nine out of eleven parameters studied.

Nisar *et al.* (2022) examined F₂ populations, derived from four parents of upland cotton, i.e. CRIS-134, VH-329, IUB-222, CIM-240 which were sown in the Plant Breeding and Genetics section, University of Agriculture, Faisalabad; Pakistan, during the normal growing season of cotton. A Randomized Complete Block design with three replications was used for the study. Plant height (PH), boll weight (BW), bolls/plant (BP), hundred seed weight (100SW), yield/plant (YP), ginning out turn (GOT percent), fiber fineness (FF), fiber length (FL), and fiber strength (FS) all revealed substantial variation in the analysis of variance (FS). It was investigated from the data that the Genotype CIM-240×CRIS-134 had maximum yield and genotype CRIS-134×IUB-222 had maximum fiber strength proved as best for the selection in future breeding.

Saraswat *et al.* (2022) characterized 50 elite lines of cotton during kharif 2021 at CCSHAU, Kaul for better documentation and utilization in breeding programmes. Significant differences were observed among cotton genotypes for morphological and

fiber quality traits. Values of treatments mean sum of squares were large as compared to error mean sum of squares for all the traits. Values of critical differences varied from 0.003 to 14.55 for Tannin and seed cotton per plants. Plant height of genotypes expressed variation from 123 to 225 while growth habit showed 46.4 to 76 values. Maximum divergence observed for cotton seed per plant as values deviated from 65.4 to 234.4 whereas seed weight per boll registered change from 3.2 to 4.9. Values of seed index exhibited change from 44 to 8.9 and number of bolls per plant maintained deviation from 15.6 to 55.3 while monopods per plant expressed variation from 1.2 to 3.6 only among the genotypes. Marginal variation from 3.0 to 5.0 exhibited by number of locules per boll along with less changes in seeds per locule as 5.0 to 7.6 only. Days to first flower expressed changes from 51.6 to 66.6. Ginning (%) ranged from 19.6 to 43.3 while lint index deviated from 1.6 to 6.4 in this set of cotton genotypes.

Seyed *et al.* (2022a) conducted a study in the Moghan Agricultural Research Station affiliated with the Ardabil Agricultural and Natural Resources Research and Training Center, Iran. In this study, 35 cultivars of cotton were evaluated for yield and fiber related traits during two consecutive years. They found that all the traits were significantly affected by plots and cultivars over two years. Among the cultivars, the highest number of the bolls, boll weight, first picking yield, total yield, number of fiber strength, fiber traction, and fiber percentage were 24.5 g, 6.30 g, 3454 kg/ha, 5431 kg/ha 34.5 g/tex, 7.30%, and 45.80% observed in cultivar Shayan, while the second picking yield, earliness percentage and fiber fineness were 2323.3 kg/ha, 73.5% and 4.55 µg/inch obtained in cultivars Varamin, Avangard and Armaghan, respectively.

Seyyed *et al.* (2022b) conducted a study in the Moghan Agricultural Research Station affiliated with the Ardabil Agricultural and Natural Resources Research and Training Center, Iran. In this study, 35 cultivars of cotton were evaluated for yield and fiber related traits during two consecutive years. They found that all the traits were significantly affected by plots and cultivars over two years. Among the cultivars, the highest number of the bolls, boll weight, first picking yield, total yield, number of fiber strength, fiber traction, and fiber percentage were 24.5 g, 6.30 g, 3454 kg/ha, 5431 kg/ha 34.5 g/tex, 7.30%, and 45.80% observed in cultivar Shayan, while the second picking yield, earliness percentage and fiber fineness were 2323.3 kg/ha, 73.5% and 4.55 µg/inch obtained in cultivars Varamin, Avangard and Armaghan, respectively.

Sahar *et al.* (2021) performed a study where analysis of variance exhibited significant variation for all studied traits except total number of nodes and the height to node ratio.

Sarwar *et al.* (2021a) carried out a study at Cotton Research Station, Ayub Agricultural Research Institute, Faisalabad, Pakistan during 2019 to estimate the genetic variation in 6 genotypes of cotton at three different sowing dates regarding cotton yield traits. The ANOVA showed that varietal effects were significant for all studied traits i.e. fiber length, fiber fineness, yield except ginning out turn GOT (%).

Hayat & Bardak (2020) screened a diverse germplasm at East Mediterranean Transitional Zone Agricultural Research Institute of Kahramanmaras and at farmer field in Diyarbakir Turkey during 2016 to assess variation for ginning outturn and fiber quality. The genepool was consisted of global collections namely Acala Maxa, Fibermax810 and Stoneville-453 derived from USA, Delcerro (Venezuela), Carmen (Australia), Taskent-6, Samarkand Uzbek (Uzbekistan), Aleppo-40 (Syria), Albania-6172 (Albania), Lachata, Nata (Spain), NSCH777 (India) and elite cultivars from Turkey including Carla, Nazilli84S, Ozbek142 and Fantom. Analysis of variance showed highly significant differences among genotypes for all traits on pooled data basis.

Bradow & Davidonis (2020) opined that, ideal cotton (*Gossypium sp.*) fibers are said to be as white as snow, as strong as steel, as fine as silk and as long as wool. It is difficult to incorporate these specifications favored by cotton processors into a breeding program or to set them as quantitative goals for cotton producers. Since the early 1980s in the USA, the USDA-AMS cotton classing offices have become the primary connection for fiber quality between cotton producers and processors. The high volumes of cotton passing through the classing offices every year have forced workers there to make compromises for the sake of speed and productivity, and to develop rapid, semiautomatic classing techniques that have blurred some fiber-quality definitions in ways that may favor one industry segment over another. The vertical integration of the U.S. cotton industry from field to fabric depends on efficient use and cooperative refinement of the existing line of communication. Fiber-classing technologies now in use and under development and evaluation allow quantitation of fiber properties, application of improved standards for end-product quality, and, most importantly, creation of a fiber-quality language and a system of fiber-quality measurements that can be meaningful and useful to producers and processors alike. A cotton physiologist working in production research examines the

interface between cotton production and processing in terms of the fiber properties currently quantified by the USDA-AMS cotton-classing offices, describes the measurement protocols available, and investigates possible environmental sources of the significant variations in fiber quality that reduce grower and processor profits. The interaction of growth environment, genetic potential, and fiber properties quantified at harvest are discussed where appropriate data or references exist.

Bhatti *et al.* (2020a) performed a study to explore genetic mechanism of yield related traits in upland cotton using thirty diverse regional cotton varieties at Cotton Research Sub-Station Raiwind, during kharif season 2015 using complete randomize block design in three replications. Analysis of variance depicted highly significant differences for all the selected traits viz, plant height, days to first flower, number of monopodial branches, number of sympodial branches, number of bolls/plant, nodes/plant and seed cotton yield.

Bhatti *et al.* (2020b) evaluated fifteen upland cotton genotypes for their genetic potential under semi-arid conditions to characterize cotton genotypes on the basis of their performance. A significant variation was observed among all the investigated attributes.

Chapepa *et al.* (2020) determined the degree of association between seed cotton yield and important traits, through a study using an RCBD experiment with ten genotypes. Seed cotton yield, lint yield, boll weight, bolls per plant, seed weight, plant height, GOT, fibre length, elongation, fineness and strength, were collected and analysed. Analysis of variance revealed significant differences for boll weight, seed weight, plant height and GOT.

Killi & Beycioglu (2020) carried out a study to determine fiber yield parameters of 46 cotton genotypes, two of which belong to *barbadense* and forty-four of *hirsutum* species, under ecological conditions of Kahramanmaras, Turkey. Based on two-year average results, cotton cultivars BA-119 (1746.30 kg ha⁻¹), ST-468 (1737.45 kg ha⁻¹), ST-488 (1696.10 kg ha⁻¹) and Furkan (1623.15 kg ha⁻¹) gave higher fiber yield per hectare than the others. Giza-45, Furkan and Baly-308 varieties yielded fiber lengths over 30 mm, while other genotypes produced shorter fiber length values. All cotton genotypes had coarse fibers ranging from 4.32-5.35 micronaire. Is-1, Baly-308 and Urania cotton cultivars were distinguished with high fiber strength values.

Mahdi & Emam (2020) carried out a study at the experimental farm of the Faculty of Agriculture, Fayoum University, Egypt, during 2018 and 2019 seasons to assess the relationship between seed cotton yield per plant and five earliness measures in three Egyptian cotton varieties (Giza 80, Giza 90 and Giza 95). The results revealed that significant differences between varieties in all characters under study.

Modi *et al.* (2020) carried out a study to determine genetic variability through yield contributing traits as well as seed cotton yield in 26 cotton germplasm. The germplasm 170-CO2-10-1(H) recorded higher seed cotton yield per plant (86.4g), boll weight (4.20g) and plant height (143.3 cm). Thus, it shows that plant height and boll weight appeared to be the most important yield contributing characters.

Rehman *et al.* (2020) conducted a two-years research to estimate association among various yield contributing parameters of cotton, i.e., seed cotton yield, seed index, ginning outturn (GOT), fiber length, fiber strength, and fiber fineness. Association analysis revealed that seed cotton yield had a significant positive correlation with GOT, staple length and fiber strength. Staple length and fiber strength were negatively linked with each other. The parent IUB-222 was found to be the best for GOT, seed cotton yield, and seed index. The genotypes namely, NIAB-414 and VH-367 were identified as the best parents for fiber length, strength, and fineness. Among the crosses NIAB-414 × IUB-222 was the best for seed index, seed cotton yield and fiber fineness. The combination of A555×CIM-632 was the best for fiber length and strength, and VH-367 × CIM-632 proved the best for GOT.

Ali *et al.* (2019) governed an experiment conducted at Cotton Research Farm, Jagodishpur, Jessore during the season 2017-2018 to evaluate seed cotton yield, cumulative % of seed cotton yield, ginning and lint of five materials and control variety CB-14. Among them, five were encoded JA-16/1, JA-08/4, JA-08/5, JA-16/2 and JA-0510 respectively. The other was control variety CB-14. Significant difference was found among the genotypes for the traits of vegetative branches/plant, node number of first fruiting branches/plant, secondary fruiting branches/plant, days to 50% flowering, bolls/plant, un-burst bolls/plant, single boll weight (g), plant height (cm), yield (kg/ha). The line JA-08/4 produced the highest seed cotton yield (4078 kg/ha) and highest lint (1656 kg/ha) than the control variety CB-14. The line JA0510 showed the highest GOT (40.50%) percentage. Among the five entries and control variety CB-14, the highest

upper half mean length (33.47 mm), the highest strength (35.23 g/tex) value were found in the line JA-0510 and the lowest micronaire value (3.56) was found in the line JA16/1.

Araújo *et al.* (2019) evaluated upland cotton cultivar fiber quality, when subjected to water deficit periods, on the phenological stages. The experiment was carried out at the Campina Grande Federal University, Pombal county Campus, Paraíba State, Brazil, between June and December 2015. Treatments were formed using a split-plot arrangement, in which the plots were 6 water deficit periods (P) (P1 = No deficit; P2 = Deficit in the initial growth stage; P3 = Deficit in the flower bud stage; P4 = Deficit in the flower stage; P5 = Deficit in the boll stage; and P6 = Deficit in the open boll stage) and the subplots, 2 upland cotton cultivars (C) (C1 = Brazil Seeds 286 and C2 = BRS 336), in randomized block design, with 4 replicates. The water deficits applied affected cultivars fiber quality, except maturity, reflectance and yellowness. The treatment without water deficit promoted the best fiber values, except of short fiber index, elongation and micronaire. Tested upland cotton cultivars were more tolerant to water deficit in the initial growth and boll stages. In general, BRS 336 was more tolerant to water deficits than BRS 286.

Jarwar *et al.* (2019a) executed a research experiment to investigate the presence of genetic variation existing as components of variation, correlations and path-coefficient analysis of yield and fiber quality traits viz. plant height, sympodial branches per plant, bolls per plant, boll weight, seed index, seed cotton yield, ginning outturn percentage, fiber length, fiber fineness and. The research experiment was conducted comprising of 12 cotton cultivars planted under randomized complete blocked design (RCBD) at the experimental field of Cotton Research Institute Anyang Henan, China, during the cotton crop season 2017-18. The cultivars included 705230, 705240, 705242, 705244, 705252, 705360, 705366, 705368, 705398, 705441, 705482, and 705542 accessions of upland cotton (*Gossypium hirsutum* L). The results in analysis of variance revealed that plant height, sympodial branches, bolls per plant, and seed cotton yield and fiber fineness showed highly significant differences, whereas boll weight, seed index, ginning out turn percentage and fiber length exhibited significant differences. In conclusion results showed that genotype 705244 has the highest plant height whereas genotype 705244 produced a maximum number of sympodial branches per plant similarly and genotype 705244 exhibited as, as a superior variety for cotton with reference to seed production and might be commercialized as a high yielding variety.

Rathinavel (2019) characterized 320 cotton (*G. hirsutum*) germplasm accessions for distinctiveness. Under field and laboratory, 26 qualitative traits and 14 quantitative traits were recorded. There is no variation observed for boll bearing habit and boll opening. Higher coefficient of variation was recorded for vigour index, seed cotton yield/plant and fibre strength.

Sirisha *et al.* (2019) investigated 45 crosses developed by making crosses among the 10 selected genotypes having very good combining ability for quality traits in half diallel fashion. These crosses were evaluated for quality traits over the commercial checks (Bunny BT and KCH 707) for their standard heterosis before they are being considered for commercial cultivation. The cross combination, BBGH-77 x BBGH-1, expressed the highest heterotic effects for all the quality characters over the check Bunny Bt and might be considered for commercial release after confirming its superiority over locations.

Sun *et al.* (2019) evaluated 719 global collections of *G. hirsutum* L. for five fibre-related traits during two consecutive years in eight different environments. A series of phenotypic data for fibre quality traits were obtained. They found that fibre quality traits showed a wide range of variation among the *G. hirsutum* accessions over 2 years. In general, accessions from outside China tended to have higher fibre length (FL) and fibre strength (FS) than did Chinese accessions. Among different regional accessions in China, North/Northwest accessions tended to have the highest FL, FS and best fibre micronaire. By assessing five fibre quality traits over 2 years with genotypic data, 31 elite germplasms reaching double-thirty quality values ($FL \geq 30$ mm and $FS \geq 30$ cN·tex⁻¹) were selected.

Alam (2018) commanded an experiment at Cotton Research Farm, Sreepur, Gazipur during kharif season 2017-18. The 101 cotton genotypes were sown in randomized complete block design with three replications. To evaluate genotypes for earliness 15 quantitative plant traits were selected such as days to first flowering, node number of 1st fruiting branch, vegetative branches plant⁻¹, primary and secondary fruiting branches plant⁻¹, number of bolls plant⁻¹, seed cotton yield (t/h) and maturity days. The mean squares from analysis of variances showed that genotypes differed significant ($P \leq 0.01$) for all the studied traits except GOT%. The genotypes, BC-0479, BC0457, BC-0495, SR-16 and Rupali-1 were characterized as early maturing or short field duration cotton varieties.

Amin (2018) conducted an experiment at Cotton Research Center, Rangpur during the crop season 2017-18 to rejuvenate the old stock of germplasm and also to observe their agronomic performance. Twenty-four stocked cotton germplasm were taken for rejuvenation whose accession numbers were BC-0496 to BC-0512, HC-2, BC-0042/A, BC-0042/B, BC-0111, BC-0113, BC-0119 and BC-0120, respectively. About 200 to 700 g selfed seeds were obtained from each accession. The accession number BC-0497 produced highest seed cotton yield (3.96 t/ha) which was followed by BC-0501 (3.52 t/ha), BC-0042/B (3.41 t/ha) and BC-0505 (3.18 t/ha) respectively. The accession number BC-0496 shown highest number of bolls per plant (73) which was followed BC-0498 (68), BC-0042/B (65) and BC-0497 (63) respectively. The highest single boll weight (6.2 g) was found in BC-0501, BC-0503 and BC-0508 which was followed by BC-0502 (6.0 g), BC-0507 (6.0 g), BC-0499 (5.9 g) and BC-0510 (5.8), respectively. Highest ginning out turn (45.24) percentage was found in BC-0512 which was followed by BC-0511 (42.99), BC-0501 (42.27), BC-0510 (41.90) and BC-0504 (41.12) percentage respectively.

Monisha *et al.* (2018) investigated a total of fifty-four *Gosypium hirsutum* L F₁ using six lines (four compact and two non-compact) with nine non compact testers in line x tester fashion were evaluated for seed cotton yield traits. Analysis of variance revealed significant differences among the genotypes for various characters studied indicating the presence of ample variation for effective selection.

Eswari *et al.* (2017) carried out an experiment at college farm, college of Agriculture, Rajendranagar, Hyderabad, to determine genetic variability in upland cotton. The analysis of variance involving a set of 11 genotypes for eleven characters viz., days to 50 % flowering, number of monopodia/plant, number of sympodia/plant, number of bolls/plant, boll weight, seed cotton yield/plant, ginning percentage, 2.5 % span length, uniformity ratio, bundle strength and micronaire value, revealed highly significant mean sum of squares for all the characters indicating greater diversity among the genotypes.

Ahmad *et al.* (2016) evaluated two parents B557, FH153 and their F₂ population for the existence of genetic variability for the characters under study during 2014-2015, at University of Agriculture, Faisalabad. The experiment was carried out in a randomized complete block (RCB) design with three replications. Data were recorded for plant height (cm), internodal distance (cm), number of monopodia, number of sympodial, leaf

hairiness, pedicel length (cm), boll size, number of bolls per plant, boll weight (g), seed cotton yield per plant (g), ginning out turn (%), fiber fineness ($\mu\text{g}/\text{inch}$), fiber strength (g/tex) and staple length (mm). The parent FH153 had maximum mean value for plant height (96.4cm), sympodial branches (10.33), boll weight (3.02g), fiber finess (5.13 $\mu\text{g}/\text{inch}$), fiber strength (21.49g/tex) and staple length (27.53mm). Parent B557 showed maximum (1.68cm) pedicel length whereas, F_2 population had maximum (3.45cm) internodal distance, monopodial branches per plant (3.01), leaf hairiness was of profuse, medium and sparse, boll size was of small, medium and large, number of bolls per plant (24.91) and lint percentage (38.62%). The results also showed that linkage of these traits did not affect yield so might be used as selection criteria for future cotton breeding programs.

Yaqoob *et al.* (2016) evaluated two parents FH153, KZ191 and their F_2 population for the existence of inter-relationship of characters under study during 2014-2015, at University of Agriculture, Faisalabad. The experiment was carried out in a randomized complete block (RCB) design with three replications. Data were recorded for Plant height (cm), number of sympodial branches, number of monopodial branches, pedicel length (mm), number of bolls per plant, number of seed per boll, boll weight (g), seed cotton yield (g) ginning out turn (%), fiber fineness (μ/inch), staple length (mm), fiber strength (g/tex). The obtained data were analyzed through correlation analysis at 0.01 and 0.05 significance level. The traits under study showed considerable range of phenotypic variability. The parent FH153 had maximum mean value for all traits under observation except fiber strength. The F_2 population of parents (FH153×KZ191) had minimum results for all traits. The parent KZ191 showed maximum fiber strength (21.1 g/tex). The F_2 population of parents (FH153×KZ191) had minimum results for all traits. The results also showed positive association of characters with each other except staple length showed negative association with fiber strength.

Baloch *et al.* (2015) conducted a research utilizing eight promising genotypes of upland cotton for eight quantitative and qualitative traits. Results depicted significant differences ($P \leq 0.05$) among the varieties for all the eight quantitative and qualitative traits, exhibiting the availability of substantial genetic diversity among the cultivars for studied traits. Hence, these promising cultivars can further be exploited in various

breeding programs to improve various quantitative and qualitative characters in cotton genotypes.

Liaqat *et al.* (2015) investigated seventy-five genotypes of cultivated cotton (*Gossypium hirsutum* L.) for morphological and fiber quality characteristics i.e., plant height, monopodial branches, sympodial branches, boll weight, seed volume, seed density and seed index. Data were subjected to analysis of variance. ANOVA revealed highly significant variability among genotypes for all the characteristics studied.

Soomro *et al.* (2015) conducted a study to determine the early maturing genotypes by evaluating plant traits in ten advance strains of upland cotton varieties developed by Central Cotton Research Institute Sakrand for exploitation more effectively and efficiently in future breeding programs. The analysis of variance of varieties was highly significant for all the traits studied. Among the genotypes CRIS-613 proved early maturing as characters studied viz. node number of 1st sympodial branch (5.07), days taken to 1st flower (42.03), maximum number of bolls at 90 and 150 days after planting and opened highest bolls at 120 and 150 days after planting. Whilst, CRIS-342, CRIS-611, CRIS-617 and CRIS-618 produced lowest followed by node number of 1st sympodial branch, days taken to 1st flower, number of bolls setting and opening respectively. Therefore, it is suggested that CRIS-613, CRIS-611, CRIS-617, CRIS-618 and CRIS-342 are considered as early maturing genotypes. Whereas, CRIS-610, CRIS-619, CRIS-620 and CRIS-621 produced more number of nodes for 1st sympodial, more days taken to 1st flower, setting minimum numbers of bolls and open less numbers of bolls, it indicated that these are the late maturing genotypes. It was further concluded that traits viz. node number of 1st sympodial branch, days taken 1st flower, setting of bolls at 90, 120 and 150 days after planting and bolls opening at 120 and 150 days after planting were more effective and efficient indicators for selecting early maturing genotypes in upland cotton.

Abbas *et al.* (2015) conducted a study at Cotton Research Institute, Ayub Agricultural Research Institute, and Faisalabad Pakistan during 2012. The boll size of Sbe-18 was large; tapering shape, good opening and leaf was of medium size as compared to all other varieties.

Baloch *et al.* (2014) investigated eight upland cotton genotypes viz., CRIS-134, Sadori, Chandi, NIAB-78, AA-802, IR-3701, IR-1524 and FH-113 to characterize the early maturing cotton genotypes. The experiment was laid out in a randomized complete block design with four replications during 2012 at Sindh Agriculture University, Tandojam, Pakistan. The mean squares indicated significant ($p \leq 0.01$) differences among the genotypes for all the traits. The two cultivars i.e. CRIS-134 and Sadori were characterized as early maturing in terms of days to 1st flower, formed sympodial branches at relatively lower nodes, set 1st effective boll on lower sympodia, formed and opened maximum bolls at 90 and 120 days after planting (DAP) and weighed medium size bolls. For yield traits, maximum bolls plant⁻¹, bigger bolls, higher seed cotton yield and more lint percentage were obtained from cultivars CRIS-134, Sadori, IR-3701 and FH-113. Regarding fiber traits, the longer staple with desirable micronaire were recorded by CRIS-134, AA-802 and FH-113.

Ahmad *et al.* (2008) examined 13 upland cotton genotypes for studying different morphological traits related with earliness i.e., days to first flower, first sympodial branch node number, sympodial branch number with first effective boll, sympodial branch length, boll opening (%) after 120 days of planting, boll weight and seed cotton yield per plant. All the varieties differed significantly for all these traits showing a variation range of 41 to 48, 5.0 to 9.0, 5.0 to 10.3, 14.2 to 35.7 cm, 53.0 to 81.0%, 2.7 to 4.5g, 126.0 to 196.3g, respectively. The new strain VH-144 and VH-156 exhibited great yield potential for earliness as they took minimum days to first flower (40.5 and 41.4, respectively) and more boll opening percentage at 120 days after planting (81 and 80.9, respectively). It was concluded that selection for early setting of first flower, lower value of first sympodial branch with first effective boll and boll opening after 120 days of planting could lead to development of early maturing variety in cotton.

Glade *et al.* (1981) opined that cotton quality characteristics are increasingly measured by instruments rather than manually. Increased competition in the industry, consumer demand for a wider range of products, and more stringent standards for product use have prompted more intensive fiber quality examination. Over 92 percent of textile firms surveyed own one or more fiber testing instruments. Ninety percent own instruments for testing fineness and maturity, 70 percent for measuring length and length uniformity, and 60 percent for determining fiber strength. This report presented information on the

determination and use of cotton quality measures, and the development of high-volume instrument testing systems.

2.6 Review of literature on genetic parameter studies of yield and yield related traits in upland cotton

Amer & Hassan (2023) conducted an experiment at Sakha Station of Agricultural Research, over 2021 and 2022 seasons to evaluate the variance among some Egyptian cotton genotypes. The genotypes were arranged in RCBD design replicated quadric. Genotypic and phenotypic exhibited broad scope of variability for all traits. Seed and lint cotton yields had moderate PCV values, while the remainder traits showed low values, GCV showed moderate value for lint yield and low values for the remainder traits. Broad sense heritability recorded low value for boll weight to very high values for most of traits. For GS%, moderate values were recorded for seed cotton yield, lint yield and earliness index while the rest had low values. In this investigation, seed cotton yield, lint yield and earliness index exhibited high heritability together with moderate genetic advance values, subsequently, direct selection for such traits is anticipated to induce considerable improvement in next generation.

Mahdy *et al.* (2023) conducted an experiment to study the gene action that controls yield and quality traits of Egyptian cotton under normal and saline soil. Nine lines were crossed to three testers in a line $\times$ tester mating design. The 27 crosses and their parents were evaluated under normal and saline soil conditions in the seasons of 2018 and 2019 using a randomized complete block design with three replicates. Both additive ($\sigma^2 A$) and dominance ($\sigma^2 D$) variances were higher under normal soil than under salinity stress conditions for all the analyzed characteristics. Under normal soil, the ratio $\sigma^2 A / \sigma^2 D$ was less than unity for seed cotton yield/plant (SCY/p), number of bolls (NB/p), number of seeds (NS/b), boll weight (BW), lint yield (LY/p), and lint%. $\sigma^2 D$ was not significant for seed index (SI), lint index (LI), days to the first flower (DFF), plant height (PH), and upper half-mean length (UHML).

Patel *et al.* (2023) aimed to investigate genetic variability among the all genotypes along with heritability presence, genetic advance per cent over mean, correlation studies and path coefficient for 17 different characters in 16 genotypes, including sixty-four F_1 hybrids, and one check G. Cot. Hy. 18 of upland cotton (*Gossypium hirsutum* L.). The

trial was demonstrated during the Kharif season of 2022 at the Regional Research Station (RRS), Anand Agricultural University, India. Variability studies revealed high phenotypic coefficient of variation (PCV) and moderate genotypic coefficient of variation (GCV) for the number of bolls per plant, lint yield per plant and seed cotton yield per plant. Additionally, the number of monopodia per plant exhibited a high estimate of both PCV and GCV. Heritability analysis showed high heritability for various traits, including days to 50% flowering, days to 50% boll bursting, plant height, monopodia per plant, sympodia per plant, number of bolls per plant, boll weight, lint yield per plant and seed cotton yield. Moreover, days to 50% flowering, plant height, number of monopodia per plant, number of sympodia per plant, number of bolls per plant, boll weight, lint yield per plant seed index, fiber strength, and fiber fineness and seed cotton yield demonstrated both high heritability and significant genetic advance as per cent over the mean.

Divya *et al.* (2023) studied thirty genotypes of cotton to estimate genetic variability, heritability (broad sense) and genetic advance for 15 traits during kharif, 2022. PCV was greater than GCV for all the traits except days to 50% flowering. High PCV and GCV were recorded for number of monopodial branches, lint index, lint yield and ginning percentage. High heritability coupled with genetic advance as percent of mean was observed for number of monopodial branches, plant height, seed cotton yield, lint index, lint yield, ginning percentage and micronaire and indicating preponderance of additive gene action and hence simple selection would be effective for the improvement of these traits.

Khakwani *et al.* (2022) investigated five varieties namely FH-414, FH-415, FH-490, FH-492, and FH-ANMOL as lines and crossed with three testers namely NIAB- SANAM, CIM-602, and EYE-111 during the year 2019-20. Data was recorded for plant height (PH), number of monopodial branches (MB), number of sympodial branches (SB), number of bolls/plant (BP), boll weight (BW) and seed cotton yield (SCY). FH 414 proved to have very good GCA for BW. FH-415 showed good GCA for monopodial branches. Similarly, FH-492 for SB, BW, BP and SCY. FH-ANMOL proved best combiner for, SB, BP, and SCY. CIM-602 was good combiner for BP, SB, BW and SCY. EYE-111 found to be a good combiner for BP. The hybrid FH-415 $\times$ CIM-602 showed good SCA for MB and FH-ANMOL $\times$ EYE-111 for SB. For bolls/plant (BP)

FH-492 × EYE-111, for boll weight (BW) FH-ANMOL × EYE-111, and for seed cotton yield (SCY) FH-492 × EYE-111 proved to be very good hybrid. These results indicated that these lines and testers have desired variability to exploit in the traits which were investigated.

Manan *et al.* (2022) investigated eight cotton genotypes with their 15 F₁ hybrids (five lines, three testers) which were grown in the field conditions under randomized complete block design (RCBD) with two treatments i.e. normal and high- temperature stresses with two replications followed by split-plot arrangement to understand the effect of heat stress on morphology and the fiber quality of the cotton. Data were collected for yield-related parameters. Under both conditions, the number of bolls, boll weight, seed cotton yield, lint index, seed mass per boll had high heritability values along with high genetic advance percent mean which revealed, these traits were controlled by additive gene action. Under both temperature conditions, FH-458, IUB-65, CRS-2, and FH-313 were good general combiners for yield-related traits. The cross combination of IUB-013× CRS-2 and FH-458× FH-313 was good specific combiner for plant height and seed cotton yield. These identified parents and cross combinations might be used for improving the commercial varieties under high-temperature stress.

Meena *et al.* (2022a) carried out an investigation using twenty-five genotypes for quantitative and qualitative traits of cotton to estimate the genetic variability present within genotypes. The material was evaluated in randomized block design with four replications during kharif, 2019 at Agronomy Instructional Farm, S. D. Agricultural University, Sardarkrushi nagar. The high genotypic and phenotypic coefficient of variation was observed for number of bolls per plant followed by number of monopodia per plant, lint yield per plant and seed cotton yield per plant suggesting the presence of considerable amount of variability. High heritability coupled with high genetic advance as per cent of mean were observed for number of sympodia per plant and number of bolls per plant, lint yield per plant and seed cotton yield per plant indicates the role of additive genes and less environmental influence on the characters and presence of adequate heritable variation. Thus it can be concluded that selection based on the traits like number of monopodia per plant, number of sympodia per plant, number of bolls per plant and lint yield per plant might be beneficial for further crop improvement.

Mawblei *et al.* (2022) evaluated one hundred upland cotton germplasm accessions for eight traits to evaluate the per se performance, genetic variability, heritability and genetic advance as a per cent of mean. The estimation of variability indicated that the number of monopodia per plant had high GCV and PCV. High heritability along with high GAM was found in plant height, internode length, the number of monopodia per plant, the number of sympodia per plant, the number of bolls per plant and seed cotton yield per plant which suggested that additive gene activity had a significant influence in the inheritance of these traits.

Saraswat *et al.* (2022) characterized 50 elite lines of cotton during kharif 2021 at CCSHAU, Kaul for better documentation and utilization in breeding programmes. Cotton seed yield and Weight of seed cotton/boll expressed high Heritability with high genetic advance as % mean for additive gene action.

Sahar *et al.* (2021) performed a study to assess the genetic variations of yield and fiber quality traits in cotton. Analysis of variance exhibited significant variation for all studied traits except total number of nodes and the height to node ratio. The phenotypic coefficient of variation was higher than the genotypic coefficient of variation for all studied traits. Plant height, monopodial branches, total number of bolls, lint index, seed index, and seed cotton yield displayed high heritability in a broad sense with maximum genetic advance.

Bhatti *et al.* (2020a) performed a study to explore genetic mechanism of yield related traits in upland cotton using thirty diverse regional cotton varieties at Cotton Research Sub-Station Raiwind, during kharif season 2015 using complete randomize block design in three replications. The traits were, plant height, days to first flower, number of monopodial branches, number of sympodial branches, number of bolls/plant, nodes/plant and seed cotton yield. All the traits revealed the phenotypic variance was slightly higher than genotypic variance; similarly, phenotypic coefficient of variance was greater than genotypic coefficient of variance which also predicts presence of additive genes for these traits. The Broad sense heritability (h^2_b) estimates were observed high for all the traits under study, but in only four characters i.e. number of monopodial branches, number of sympodial branches, seed cotton yield and number of bolls/plants it coupled with high genetic gain also, which promised the further effective selection on phenotypic basis, while for other attributes indirect selection will be suitable.

Rehman *et al.* (2020) conducted a two-years research to estimate heritability among various yield contributing parameters of cotton, i.e., plant height, number of bolls per plant, number of sympodial branches per plant, seed cotton yield and boll weight. Estimates of heritability were high for all of the traits except the number of sympodial branches per plant and boll weight. The parent IUB-222 was found to be the best for plant height, the number of bolls per plant, boll weight and seed cotton yield. Among the crosses NIAB-414 $\times$ IUB-222 was the best for the number of bolls per plant and seed cotton yield, whereas, the cross of NIAB-414 $\times$ CIM-632 was good for plant height. The combination of A555 $\times$ CIM-632 was the best for the number of sympodial branches per plant, boll weight.

Jarwar *et al.* (2019a) executed a research experiment to investigate the presence of genetic variation existing as components of variation of yield traits viz. plant height, sympodial branches per plant, bolls per plant, boll weight, seed index, and seed cotton yield. The research experiment was conducted comprising of 12 cotton cultivars planted under randomized complete blocked design (RCBD) at the experimental field of Cotton Research Institute Anyang Henan, China, during the cotton crop season 2017-18. The cultivars included 705230, 705240, 705242, 705244, 705252, 705360, 705366, 705368, 705398, 705441, 705482, and 705542 accessions of upland cotton (*Gossypium hirsutum* L.). High heritability with moderate genetic advance for the number of sympodial branches, Number of bolls per plant, and seed cotton yield provide a sound ground for improvement in these through selection.

Shahzad *et al.* (2019) adopted North Carolina II mating design to cross 5 female parents with 6 male parents to produce 30 intraspecific F₁ cotton hybrids. All plant materials were tested in three different ecological regions of China during the year of 2016-2017. Additive-dominance-environment (ADE) genetic model was used to estimate the genetic effects. Results showed that yield traits except lint percentage were mainly controlled by genetic and environment interaction effects, whereas lint percentage was determined by main genetic effects. Moreover, dominant and additive environment interaction effects had more influence on yield traits. Broad-sense and its interaction heritability were significant for all yield traits. Narrow-sense and its interaction heritability were non-significant for boll number and seed cotton yield. Results of the study provided important information about genetic control of yield. Further, this study identified that

parental lines, e.g., SJ48-1, ZB-1, 851-2, and DT-8 could be utilized to improve yield traits in cotton.

Monisha *et al.* (2018) investigated a total of fifty-four *Gosypium hirsutum* L F₁ using six lines (four compact and two non-compact) with nine non compact testers in line x tester fashion were evaluated for seed cotton yield traits. The phenotypic coefficient of variation (PCV) was slightly higher in magnitude than genotypic coefficient of variation (GCV) for all characters indicating the influence of environment over the expression of the traits studied. High phenotypic and genotypic coefficients of variation were obtained for number of monopodial branches per plant and single plant yield. High heritability coupled with high genetic advance (as per cent of mean) was observed for characters viz., number of monopodial branches per plant, number of sympodial branches per plant, number of bolls per plant, single plant yield and internode length.

Eswari *et al.* (2017) carried out an experiment at college farm, college of Agriculture, Rajendranagar, Hyderabad, to determine genetic variability, heritability and genetic advance estimates in upland cotton. The value of phenotypic coefficient of variation (PCV) is greater than genotypic coefficient of variation (GCV); it means that the apparent variation is not only due to genotypes but also due to influence of environment. High GCV and PCV were observed for number of monopodia per plant. The difference between the estimates of PCV and GCV were low for all the characters except number of monopodia per plant thereby indicating that lesser role played by the environment in the expression of these characters. The character's number of bolls/plant, boll weight, days to 50 % flowering, bundle strength, ginning percentage, 2.5 % span length, and seed cotton yield/plant exhibited high estimates of heritability. High heritability coupled with high genetic advance as percent of mean was observed for number of bolls/plant, boll weight and Seed cotton yield/plant.

Farooq *et al.* (2015) designed a research work to estimate genotypic analysis between seed cotton yield and yield contributing traits in 18 cotton cultivars. The material was sown on 15th of May to observe association among yield and its related traits. Heritability was estimated to determine the selection criteria under normal growing conditions. The values of phenotypic coefficient of variation (PCV%) were higher in magnitude than the genotypic coefficient of variation (GCV%) for all the traits. Heritability (broad sense) revealed higher estimates for all traits like plant height (98%), nodes to first fruiting

branch (95%), bolls per plant (91.5%), sympodia per plant (88.8%), yield (88%) monopodia per plant (84.7%) and for boll weight (82.3%).

Liaquat *et al.* (2015) investigated seventy-five genotypes of cultivated cotton (*Gossypium hirsutum* L.) for morphological characteristics i.e plant height, monopodial branches, sympodial branches, boll weight, seed volume, seed density and seed index. Estimates were made for genetic advance, broad sense heritability and coefficient of variance for the traits. The estimates for heritability were higher for seed index (0.93) and plant height (0.93). The highest value (6.4) for genetic advance was observed for sympodial branches whereas lowest value was (0.17) for boll weight.

Pujer *et al.* (2014) evaluated sixty-eight diverse genotypes of American cotton (*Gossypium hirsutum* L.) for 13 quantitative fibre quality traits. The variability studies indicated that high PCV and GCV was observed in case of seed cotton yield/ plant and number of bolls/plant while moderate PCV and GCV was observed in case of days to first flower, plant height and boll weight. Seed cotton yield /plant, days to first flower, plant height, number of bolls/plant and boll weight showed high heritability with high genetic advance over mean.

Abbas *et al.* (2015) conducted a study at Cotton Research Institute, Ayub Agricultural Research Institute, and Faisalabad Pakistan during 2012. It was concluded that higher heritability and genetic advance was found for monopodial and sympodial branches, nodes to first flower days, number of bolls and plant height. Higher heritability and genetic advance indicated that selection might be helpful for the improvement of yield and quality of cotton.

Baloch *et al.* (2015) estimated heritability in eight promising genotypes of upland cotton for eight quantitative and qualitative traits. High heritability estimates were found for all studied traits, with the exception of monopodial branches plant⁻¹, indicating that these traits were primarily controlled by additive gene effects and direct selection may be proved useful for these traits.

Dinakaran *et al.* (2012) conducted a study to assess the salt tolerance of 32 popular upland varieties released for general cultivation between 1980 and 2001 in India. The study was carried out in normal as well as saline-alkaline condition; in which salinity

were created using bore well water irrigation and the average electrical conductivity level of bore well water is 3.10 ds/m. The 32 upland cotton genotypes under both salinity and normal conditions revealed high GCV and genetic gain for 2.5 per cent span length, number of bolls per plant, boll weight, lint yield per plant, leaf area index, and seed cotton yield and these traits might be improved by simple selection.

Song *et al.* (2012) evaluated inheritance and interrelationship of phenotype and genotype of earliness traits in a diallel analysis involving six early-maturing parents. Date of first square (DFS), date of first flower (DFF), date of first open boll (DFOB), number of node first sympodial branch (NNFSB), and harvested rate before frost (HRBF) as earliness traits of six parents, 15 F₁ hybrids and 15 F₂ progenies were investigated from 2005 to 2008. The experiment design was a randomized complete block design with three replications. Additive, dominance and epistasis effects were analyzed with ADAA (additive dominance-epistasis) model. HRBF, DFF, and DFOB showed significant additive genetic variances. Heritability estimates ranged from 0.088 (HN, narrow sense) and 0.416 (HNE, environment interaction) for HRBF, to 0.103 (HN) and 0.524 (HNE) for DFF, and to 0.187 (HN and 0.519 (HNE) for DFOB. Dominance genetic effects for DFS, DFF, DFOB, and NNSFB were stronger than additive effects. Additive-by-additive epistatic effects for DFS, DFOB, and NNSFB were detected and affected by environment. Heredity of earliness traits among parents and their hybrids were also detected and parents A1, A2, B1, B2, and B3 might be used to improve earliness traits of short season cotton cultivars.

2.7 Review of literature on character associations of different traits in upland cotton

Beegum *et al.* (2025) conducted a study to analyze fiber quality data from 40 upland cotton cultivars, including TM1. The measured fiber quality from all cultivars was then compared with the model simulated fiber quality. The Pearson correlation coefficient (*r*) between the measured and simulated values was negative for all fiber quality traits, and Wilmotts's index of agreement (WIA) was below 0.45, indicating a strong model bias toward TM1 without incorporating cultivar-specific parameters. After incorporating cultivar-specific parameters, the model's performance improved significantly, with an average *r*-value of 0.84 and WIA of 0.88.

Kanta *et al.* (2025) conducted a study with 56 genotypes of cotton in alpha lattice design with two replications at Regional Agricultural Research Station, Lam Farm, Guntur during Kharif season 2024. In this study, correlation and path coefficient analysis were assessed for 19 characters among 56 genotypes of upland cotton. Correlation studies revealed that no. of sympodia per plant, no. of bolls per plant, boll weight, seed index, lint index, seed cotton yield per plant at 140 days after sowing, seed cotton yield per plant at 165 days after sowing and oil content recorded significant positive association with total seed cotton yield per plant indicating more emphasis should be given to these traits during breeding programme to make selection effective. Further partitioning of correlation into direct and indirect effects showed that characters no. of sympodia per plant, no. of bolls per plant, lint index, seed cotton yield per plant at 140 days after sowing, seed cotton yield per plant at 165 days after sowing, tenacity value had direct positive effect on total seed cotton yield per plant. Thus, correlation and path analysis clearly indicated that direct selection based on no. of sympodia per plant, no. of bolls per plant, lint index, seed cotton yield per plant at 140 days after sowing, seed cotton yield per plant at 165 days after sowing might be helpful in developing high yielding varieties in upland cotton.

Bilal *et al.* (2024a) carried out a study to discover varied parents and assess their cross performance. The findings showed a substantial positive connection (0.81) between green bolls, plant height (0.57), and open bolls, as well as a significant positive correlation (0.52) between open bolls. On the other hand, a highly significant negative connection was observed between the number of open bolls (-0.59), number of green bolls (-0.72), plant height (-0.78), and major stem nodes (-0.70) for the cotton leaf curl viral disease.

Çakmak *et al.* (2024) showed significantly negative correlations between seed cotton yield and fiber length; ginning out-turn and fiber fineness. The decision tree model showed that fiber fineness (4.60 mic.) within commercial limits depends on the optimization of seed cotton yield per plant of approximately 54.27 g and ginning out-turn of 42.60% in single plant selection.

Chapara *et al.* (2024) executed a study during Kharif, 2022 to evaluate the character association and genetic variability of yield and its associated traits in cotton. Correlation studies revealed that seed cotton yield exhibited a substantial positive kinship with the

number of bolls per plant, the number of sympodial branches per plant, seed index, ginning out turn (GOT), and lint yield.

Gowda *et al.* (2024) evaluated an elite diversity panel of 348 upland cotton accessions in 10 environments across the US Cotton Belt of six fiber quality traits viz, upper half mean length (UHML: mm), fiber strength (FS: g tex⁻¹), fiber uniformity (FU: %), fiber elongation (FE: %), micronaire (MIC) and short fiber content (SFC: %). UHML, FS and FU were all positively correlated with each other and negatively correlated with FE, MIC and SFC.

Kadam *et al.* (2024) investigated fifty diverse elite cotton genotypes, including four check varieties, cultivated in a Randomized Block Design with two replications during the 2022-23 kharif season at the Cotton Research Station, Nanded. The study aimed to analyze correlations and conduct path analysis of yield and fibre quality traits in American cotton (*Gossypium hirsutum* L.). Correlation analysis revealed significant positive genotypic and phenotypic correlations between plant height, number of bolls per plant, boll weight, seed cotton yield per plant, fibre fineness, and seed cotton yield per plant. Path coefficient analysis determined direct and indirect trait effects on seed cotton yield per plant, emphasizing traits such as the number of bolls per plant, plant height, ginning percentage, lint index, uniformity ratio, and fibre strength which directly and positively impacted yield. Consequently, these attributes merit priority in selecting high-yielding American cotton genotypes.

Jogender *et al.* (2023) inspected 50 genotypes of *Gossypium arboreum* grown during Kharif 2021-22 under rainfed conditions at Research Area of Cotton Section, Department of Genetics and Plant Breeding, CCSHAU, Hisar with the objective to determine correlation coefficient and path analysis. Randomized block design was used with three replications and each genotype was grown in single row of 6 m length with a spacing of 67.5 × 30 cm. These genotypes were evaluated for eight morphological. The correlation studies revealed that seed cotton yield per plant showed highly significant and positive correlation with plant height, number of bolls per plant, boll weight. Furthermore, Path analysis indicated that maximum value for direct effects were observed for number of bolls per plant followed by boll weight, number of monopods per plant. Therefore, selection might be done on the basis of these studied traits for further enhancement of seed cotton yield in future cotton breeding programme.

Patel *et al.* (2023) aimed to investigate genetic variability among the all genotypes along with correlation studies and path coefficient for 17 different characters in 16 genotypes, including sixty-four F₁ hybrids, and one check G. Cot. Hy. 18 of upland cotton (*Gossypium hirsutum* L.). The trial was demonstrated during the Kharif season of 2022 at the Regional Research Station (RRS), Anand Agricultural University, India. During the correlation study, it was found that seed cotton yield per plant showed noteworthy positive correlations with several traits: the number of sympodia per plant (0.29), the number of bolls per plant (0.77), fiber length (0.26), uniformity index (0.37), lint yield per plant (0.95), fiber length (0.26) and uniformity index (0.37). Path analysis revealed that seed index and ginning outturn had the highest positive and direct effects on seed cotton yield per plant.

Subalakhshmi *et al.* (2023) investigated eight parents and 56 hybrids were evaluated for 18 economically significant traits. The study was conducted at Department of Cotton, Tamil Nadu Agricultural University, Coimbatore, in 2021-2022. The result of correlation revealed that the traits viz., number of bolls per plant, boll weight, number of seeds per boll, upper half mean length and fibre strength had a high magnitude of positive correlation with seed cotton yield at both genotypic and phenotypic levels. Path analysis represented the direct effects of upper half mean length, uniformity index, number of bolls per plant, number of seeds per boll and fibre strength on seed cotton yield. Hence, these traits might be used as selection criteria for yield improvement in cotton.

Subhashini *et al.* (2023) studied the relationship among the yield and yield contributing traits. The observations were taken in the F₁ population of 35 hybrids along with lines (7 lines) and testers (5 testers) for the traits viz. plant height (cm), number of sympodial branches per plant, the number of bolls per plant, boll weight (g), lint index (g), single cotton yield per plant (g), The results revealed that seed cotton yield per plant had strong and positive association with the traits namely number of sympodial branches per plant (0.646), number of bolls (0.633) and boll weight (0.652). Path analysis study revealed that number of sympodial branches per plant (1.5396) and boll weight (0.6285) had high direct positive effects on single plant yield and indirect very high positive effects through the traits viz., number of sympodial branches per plant via., number of bolls per plant (1.5125) and boll weight (1.0121). Hence selection based on these traits that are

positively associated and direct positive effect with the yield might produce the best outcome in the genotypes in further breeding programmes.

Vishnuvardhan *et al.* (2023) evaluated 17 cotton genotypes with a view to study of genetic parameters for different yield and yield parameters. The findings of correlation coefficient studies revealed that seed cotton yield established strong positive correlation with lint yield (0.9827) followed by plant height (0.6405) and number of bolls per plant (0.4717).

Jangid *et al.* (2022) investigated 30 elite genotypes of desi cotton as experimental material. The genotypes were sown at the Cotton Research Station in Sirsa. Correlation coefficient analysis indicated a highly significant positive relationship between seed cotton yield and the yield contributing traits. Lint yield, number of bolls per plant and boll weight had the most positive and significant correlation with seed cotton yield. The path-coefficient study revealed that the number of bolls per plant, boll weight, and lint yield was important since they had a positive direct influence on seed cotton yield per plant. As a result, selection based on the above-mentioned traits would result in genotypes with high seed cotton yield.

Mawblei *et al.* (2022) evaluated one hundred upland cotton germplasm accessions for eight traits to evaluate the per se performance, correlation and path analysis. Correlation analysis showed that the number of sympodia per plant, the number of bolls per plant and boll weight have a significant high positive correlation on the seed cotton yield. Path analysis showed that the number of bolls per plant and boll weight had the highest positive direct effect on seed cotton yield per plant.

Nisar *et al.* (2022) examined F_2 populations, derived from four parents of upland cotton, i.e. CRIS-134, VH-329, IUB-222, CIM-240 which were sown in the Plant Breeding and Genetics section, University of Agriculture, Faisalabad; Pakistan, during the normal growing season of cotton. A Randomized Complete Block design with three replications was used for the study. The goal of this research was to quantify variability and conduct a correlation analysis to see potential for selection in the F_2 population. Plant height (cm) was found to have a positive relationship with boll weight, bolls/plant, fiber strength, fiber length, and fiber fineness when the correlation coefficient was calculated. The bolls/ plant revealed a positive correlation with the boll weight and the production of

seed cotton. Boll weight revealed a positive correlation with fiber length and ginning outturn. Fiber length was positively associated with fiber strength. Correlation analysis showed that 100 seeds (g) weight was positively linked to fiber fineness, yield per plant (g) and GOT.

Sahar *et al.* (2021) performed a study to analyze the associations present in the yield and fiber quality traits in cotton. Correlation analysis revealed that seed cotton yield had a significant positive association with plant height, the number of monopodial branches, the number of sympodial branches, ginning outturn (GOT), the number of bolls, seed per boll, seed index, uniformity index, the number of sympodial branches, reflectance, and seed index at the genotypic level while a significant positive relationship was observed with plant height, the number of sympodial branches, boll number, and GOT. Plant height, monopodial branches, GOT, boll weight, seeds per boll, and short fiber index exerted direct positive effects on seed cotton yield.

Sainath *et al.* (2022) investigated 390 Recombinant inbred lines along with 6 checks which were evaluated in augmented design. The experiment was conducted in botanical garden, University of Agricultural Sciences, Dharwad during kharif 2021. The objective of this study was to improve overall productivity through use the recombinant inbred lines. The correlation study revealed that seed cotton yield was significantly and positively correlated with number of sympodial branches plants^{-1} , number of bolls plant^{-1} , boll weight, lint yield plant^{-1} , ginning outturn, seed index and lint index. Path coefficient analysis revealed that plant height, total number of monopodial branches plant^{-1} , number of sympodial branches plant^{-1} , boll weight, seed index and lint yield plant^{-1} had a favourable direct effect on seed cotton output, indicating that selecting for these traits would be quite successful in enhancing seed cotton yield in cotton.

Sarwar *et al.* (2021a) carried out a study at Cotton Research Station, Ayub Agricultural Research Institute, Faisalabad, Pakistan during 2019 to estimate the genetic variation in 6 genotypes of cotton at three different sowing dates regarding cotton quality and yield traits. The analysis of correlation depicted that days to 1st boll open had significant negative correlation with sympodial branches count, bolls count, GOT (%), fiber length and yield. However, GOT (%) and fiber length had positive correlation with yield.

Sarwar *et al.* (2021b) performed correlation in 25 cotton genotypes. Correlation analysis revealed that days to first square, flower and boll opening were positively associated

with seed cotton yield. On the other hand, staple length was negatively associated with fiber fineness and yield, respectively

Bhatti *et al.* (2020a) performed a study to explore genetic mechanism of yield related traits in upland cotton using thirty diverse regional cotton varieties at Cotton Research Sub-Station Raiwind, during kharif season 2015 using complete randomize block design in three replications. The traits were, plant height, days to first flower, number of monopodial branches, number of sympodial branches, number of bolls/plant, nodes/plant and seed cotton yield. Seed cotton yield exhibited highly significant and positive relation with sympodial branches/plant and bolls/plant whereas it displayed negative association with other studied attributes.

Bhatti *et al.* (2020b) evaluated fifteen upland cotton genotypes for their genetic potential under semi-arid conditions to characterize cotton genotypes on the basis of their performance. The correlation matrix revealed the positive association of sympodial branches per plant (SBP), number of bolls per plant (BPP) and ginning out turn (GOT) with seed cotton yield (SCY), whereas staple length (S/L) revealed positive interaction with ginning out turn (GOT) but indicated negative relationship with other yield related traits.

Chapepa *et al.* (2020) determined the degree of association between seed cotton yield and important traits, through a study using an RCBD experiment with ten genotypes. Seed cotton yield, lint yield, GOT, fibre length, elongation, fineness and strength, boll weight, bolls per plant, seed weight and plant height data were collected and analysed. Seed cotton yield was correlated with lint yield ($r = 0.71^{***}$), bolls per plant (0.27^{***}), seed weight ($r = 0.22^{***}$), fibre elongation ($r = 0.54^{***}$), strength ($r = 0.21^{***}$) and fineness ($r = 0.13^*$) at genotypic level. Boll weight was correlated with seed weight ($r = 0.56^{***}$). Ginning outturn was correlated with lint yield ($r = 0.70^{***}$), elongation ($r = 0.60^{***}$) and strength ($r = 0.50^{***}$). The adjusted R Square (0.98), low standard error (0.12) and low residual effect ($R = 0.01$) in regression analysis indicated that variability of seed cotton yield was explained by the causal variables. Lint yield had the highest direct effect on seed cotton yield ($r = 1.055$). Traits that could be used for indirect selection were bolls per plant ($r = 0.006$), seed weight (0.022) and or plant height (0.012). Ginning outturn (0.737) had the highest indirect contribution to seed cotton yield

through lint yield. Ginning outturn and fibre strength might be used indirectly to improve seed cotton yield through other traits.

Hayat & Bardak (2020) screened a diverse germplasm at East Mediterranean Transitional Zone Agricultural Research Institute of Kahramanmaraş and at farmer field in Diyarbakir Turkey during 2016 to assess variation for ginning outturn and fiber quality. The genepool was consisted of global collections namely Acala Maxa, Fibermax810 and Stoneville-453 derived from USA, Delcerro (Venezuela), Carmen (Australia), Taskent-6, Samarkand Uzbek (Uzbekistan), Aleppo-40 (Syria), Albania-6172 (Albania), Lachata, Nata (Spain), NSCH777 (India) and elite cultivars from Turkey including Carla, Nazilli84S, Ozbek142 and Fantom. Correlation analysis showed significant relationship among fiber traits such as fiber length, fiber strength and micronaire. It was concluded that some genotypes can be used as potential parents in breeding such as AB-80 and BA440 for increasing lint percentage, YB-230 and Flora for fiber length, and Delcerro and Menderes for fiber strength.

Kumar (2020) studied eleven intra *hirsutum* F₄ populations in winter rainfed condition in Cotton Research Station, Veppanthattai during the year 2017. Single plants were selected based on biometrical traits seed cotton yield (g/plant), plant height (cm), number of monopodial branches per plant, number of sympodial branches per plant, number of bolls per plant. Among the eleven crosses the two crosses viz., F 2617 x Surabhi and African I -2 x TCH 1705 -250 in which number of plants selected were higher is used to study the interrelationship. The results revealed that the positive significant correlation for the plant height with seed cotton yield per plant and number of sympodial branches per plant. Path analysis revealed the highest positive direct effects for plant height and number of bolls per plant in both the crosses. The positive indirect effect for seed cotton yield was observed for number of bolls per plant. Hence selection based on the above traits might be useful in selection programme in rainfed conditions.

Mahdi & Emam (2020) carried out a study at the experimental farm of the Faculty of Agriculture, Fayoum University, Egypt, during 2018 and 2019 seasons to assess the relationship between seed cotton yield per plant and five earliness measures in three Egyptian cotton varieties (Giza 80, Giza 90 and Giza 95). The correlation analysis indicated that earliness index and production rate index were had a high correlation and positive with seed cotton yield per plant, while days to the first flower appearance, days

to the first boll opening and mean maturity date showed negative correlation with seed cotton yield per plant. The path coefficient analysis showed that earliness index had a high direct effect (43.21%) on seed cotton yield per plant followed by production rate index (27.11%) and their interaction effect was 9.23%. The total contribution of all characters under study was 96.81%. From the above it is clear that earliness index and production rate index were the maximum traits contribution in seed cotton yield per plant therefore, might be used as useful criteria to increase cotton yield.

Rehman *et al.* (2020) conducted a two-years research to estimate association among various yield contributing parameters of cotton, i.e., plant height, number of bolls per plant, number of sympodial branches per plant, seed cotton yield and boll weight. Association analysis revealed that seed cotton yield had a significant positive correlation with plant height, number of bolls per plant and number of sympodial branches per plant. The parent IUB-222 was found to be the best for plant height, the number of bolls per plant, boll weight and seed cotton yield. Among the crosses NIAB-414 $\times$ IUB-222 was the best for the number of bolls per plant and seed cotton yield, whereas, the cross of NIAB-414 $\times$ CIM-632 was good for plant height. The combination of A555 $\times$ CIM-632 was the best for the number of sympodial branches per plant and boll weight.

Rathinavel (2019) characterized 320 cotton (*G. hirsutum*) germplasm accessions for distinctiveness. Under field and laboratory, 26 qualitative traits and 14 quantitative traits were recorded. In the Pearson's correlation, the number of bolls per plant, number of sympodia, seed cotton yield per row showed positive significant correlation with seed cotton yield per plant. These traits might be directly used as selection criteria for yield improvement in cotton.

Shahzad *et al.* (2019) adopted North Carolina II mating design to cross 5 female parents with 6 male parents to produce 30 intraspecific F₁ cotton hybrids. All plant materials were tested in three different ecological regions of China during the year of 2016–2017. Correlation analysis indicated that seed cotton yield had significant positive correlation with other yield and fiber quality attributes. All fiber quality traits had significant positive correlation with each other except micronaire. Results of the study provided important information about genetic control of fiber quality traits. Further, this study identified that parental lines, e.g., SJ48-1, ZB-1, 851-2, and DT-8 could be utilized to improve yield and fiber quality traits in cotton.

Monisha *et al.* (2018) investigated a total of fifty-four *Gosypium hirsutum* L. F₁ using six lines (four compact and two non-compact) with nine non compact testers in line x tester fashion were evaluated for seed cotton yield traits. Correlation studies revealed that seed cotton yield exhibited positive and highly significant correlation with number of sympodial branches per plant, number of bolls per plant and boll weight, lint index, ginning out turn and 2.5 per cent span length.

Isong *et al.* (2017) studied the relationship of yield components conducted on sixty hybrids obtained from a cross of ten *G. hirsutum* and six *G. barbadense* in a Line x Tester fashion. The study revealed that most of the components were positively correlated amongst each other both genotypically and phenotypically. Number of bolls per plant, Number of sympodial branches per plant and boll weight had positive and high significant correlations with seed cotton yield per plant. In all cases phenotypic coefficient was greater than genotypic coefficient suggesting environmental interference. Partitioning the association into direct and indirect effects indicated that seed index had the highest direct effect on seed cotton yield. For indirect selection, seed index, number of monopodia, number of seeds/boll will be more reliable and 2.5% span length, bundle strength, elongation percent and micronaire also are more reliable for indirect selection for fibre quality in cotton.

Ahmad *et al.* (2016) evaluated two parents B557, FH153 and their F₂ population for the existence of genetic variability for the characters under study during 2014-2015, at University of Agriculture, Faisalabad. The experiment was carried out in a randomized complete block (RCB) design with three replications. Data were recorded for Plant height (cm), Internodal distance (cm), Number of monopodia, Number of sympodial, Leaf hairiness, Pedicel length (cm), Boll size, Number of bolls per plant, Boll weight (g), and Seed cotton yield per plant (g). The obtained data were analyzed through correlation analysis at 0.01 and 0.05 significance level. The traits under study showed considerable range of genetic variability.

Bhailume *et al.* (2016) carried out a study to determine direct and indirect effects of some characters (days to first flowering, days to 50 per cent flowering, days to maturity, plant height (cm), monopodia/plant, sympodia/plant, boll weight (g), bolls/plant, halo length (mm), 100 seed weight (g)) on seed cotton yield. The experiment was carried out with 50 genotypes of *Gossypium arboreum* L. were evaluated to study the correlation

and path effects. The correlation studies revealed that plant height, monopodia/plant, sympodia/plant, boll weight, bolls/plant and halo length showed significant and positive correlation with seed cotton yield/plant. The associations of bolls/plant and days to 50 per cent flowering with seed cotton yield/plant were stronger than remaining yield traits. Path analysis revealed positive direct effect of days to 50 per cent flowering, sympodia/plant, boll weight, bolls/plant and 100 seed weight on seed cotton yield. Bolls/plant and days to 50 per cent flowering had high positive direct effect on seed cotton yield/plant and appeared to be the most important component of yield. Direct selection for these two traits could prove advantageous for improving seed cotton yield.

Yaqoob *et al.* (2016) evaluated two parents FH153, KZ191 and their F₂ population for the existence of inter-relationship of characters under study during 2014-2015, at University of Agriculture, Faisalabad. The experiment was carried out in a randomized complete block (RCB) design with three replications. Data were recorded for Plant height (cm), number of sympodial branches, number of monopodial branches, pedicel length (mm), number of bolls per plant, number of seed per boll, boll weight (g), ginning out turn (%), fiber fineness (μ /inch), staple length (mm), fiber strength (g/tex) and seed cotton yield (g). The obtained data were analyzed through correlation analysis at 0.01 and 0.05 significance level. The traits under study showed considerable range of phenotypic variability. The results also showed positive association of characters with each other except staple length showed negative association with fiber strength.

Ahsan *et al.* (2015) conducted an experiment to determine the variation and correlation of different traits with seed cotton yield per plant in *Gossypium hirsutum* accessions, at central cotton research Institute Sakrand, Pakistan during 2014. It is clear from the analysis of variance that accessions were highly significant for all the recorded traits. It was clear from the analysis that seed cotton yield was positive correlate with the most of the traits under observation. Beside lint percentage, seed cotton yield is the ultimate objective of the growing cotton. Highest seed cotton yield per plant (125.42 g) was observed in CRIS-667 followed by CRIS-665 i.e. 120 g. This type of correlation is rarely found and ultra-desirable by the cotton breeders.

Baloch *et al.* (2015) estimated correlation eight promising genotypes of upland cotton for eight quantitative and qualitative traits. Correlation analysis showed that sympodial branches plant⁻¹, boll weight and bolls plant⁻¹ made significant and positive associations

with seed cotton yield plant⁻¹. Thus, selection for these traits will ultimately enhance the chances to increase seed cotton yield plant⁻¹.

Farooq *et al.* (2015) designed a research work to estimate genotypic and phenotypic correlation coefficients and path analysis at the genotypic level between seed cotton yield and yield contributing traits in 18 cotton cultivars. The material was sown on 15th of May to observe association among yield and its related traits. The results of phenotypic and genotypic correlation showed that sympodia per plant showed positive significant correlation with boll per plant, boll weight and yield at both genotypic and phenotypic levels. Boll weight showed strong association with the yield at both levels. Path coefficient analysis revealed that all traits influenced directly and positively on yield except for bolls per plant.

Liaquat *et al.* (2015) investigated seventy-five genotypes of cultivated cotton (*Gossypium hirsutum* L.) for morphological characteristics i.e. plant height, monopodial branches, sympodial branches, boll weight, seed volume, seed density and seed index. Correlation analysis revealed positive and significant for most of the parameters. In path coefficient, the number of sympodial branches, boll weight, lint index and lint weight had maximum direct and positive effect on fiber fineness of seed cotton. Whereas, the number of monopodial branches, plant height, seed index, seed volume, seed density, staple length, fiber strength and ginning out turn (G.O.T %) had direct and negative effects on fiber of seed cotton. The grouping of genotypes possessing excelled traits signified genetic potential of the germplasm for the improvement of fiber characteristics in cotton crop.

Reddy *et al.* (2015a) carried out an experiment with 55 genotypes (45 F₁ s and 10 parents) of cotton for seventeen characters in three locations i.e. Regional Agricultural Research Station, Lam, Guntur, Agricultural Research Station, Jangamaheswarapuram and Agricultural Research Station, Darsi, Andhra Pradesh to analyze correlation and path coefficient for yield and fiber quality characters in upland cotton. Analysis of pooled data from three locations showed the character association. Seed index, lint index, micronaire value (10⁻⁶ g/inch) and lint yield plant⁻¹ were found to have significant positive association with seed cotton yield plant⁻¹ at both phenotypic and genotypic levels. The path analysis indicated that seed index and lint yield plant⁻¹ (g) showed direct positive effects and significant positive correlation with seed cotton yield plant⁻¹ (g)

revealing that due weightage should be given in selection process for economic balance among these traits to get higher seed cotton yield plant⁻¹.

Reddy *et al.* (2015b) conducted an experiment on correlation and path coefficient analysis for yield and yield contributing characters in upland cotton with 63 genotypes of cotton for seventeen characters at Regional Agricultural Research Station, Lam Farm, Guntur, Andhra Pradesh. The character association analysis revealed that 2.5% span length, bundle strength and lint yield plant⁻¹ were found to have significant positive association with seed cotton yield plant⁻¹ at both phenotypic and genotypic levels. The path analysis indicated that seed index, 2.5% span length (mm) and lint yield plant⁻¹ (g) showed direct positive effects and significant positive correlation with seed cotton yield plant⁻¹ (g) revealing that due weightage should be given in selection process with more number of bolls plant⁻¹ and more boll weight and there should be economic balance among these traits to get higher seed cotton yield plant⁻¹.

Srinivas *et al.* (2015) conducted a study on correlation and path coefficient analysis for seed cotton yield and its component traits in 56 crosses involving seven lines and eight testers. The results showed that number of monopodia per plant, number of sympodia per plant, number of bolls per plant, boll weight, number of seeds per boll and 2.5% span length were positively and significantly correlated with seed cotton yield and the traits days to 50% flowering, uniformity ratio and micronaire value exhibited significant negative correlation. Similarly, path coefficient analysis revealed that number of bolls per plant and boll weight exhibited maximum direct effect on yield.

Abbas *et al.* (2015) conducted a study at Cotton Research Institute, Ayub Agricultural Research Institute, and Faisalabad, Pakistan during 2012. Significant genotypic and phenotypic correlations were found for monopodial, sympodial branches, number of bolls, staple length and fibre fineness.

Baloch *et al.* (2014) investigated eight upland cotton genotypes viz., CRIS-134, Sadori, Chandi, NIAB-78, AA-802, IR-3701, IR-1524 and FH-113 to characterize the early maturing cotton genotypes and determine the relationship among earliness, yield and fiber quality traits. The experiment was laid out in a randomized complete block design with four replications during 2012 at Sindh Agriculture University, Tandojam, Pakistan. The correlation coefficient showed that sympodial branch length, bolls formed at 120

DAP, bolls plant⁻¹ and seed index were significantly and positively correlated with seed cotton yield. The higher correlations of yield components with yield indicated their reliable use as indirect selection criteria to improve the seed cotton yield. Some important negative correlations of lint percentage with seed index, staple length and micronaire value were also recorded which indicated that a compromise level for lint percentage might be considered while improving staple length and fiber fineness in cotton.

Pujer *et al.* (2014) evaluated sixty-eight diverse genotypes of American cotton (*Gossypium hirsutum* L.) for 13 quantitative yield related traits. The correlation study revealed that seed cotton yield was found to be positively and significantly correlated with traits like days to first flower, plant height, number of monopodial branches, number of bolls/plant, whereas it had negative association with boll weight. Path analysis revealed that days to first flower, number of monopodial branches, number of bolls/plant, boll weight, seed index, lint index, ginning out turn and uniformity ratio showed positive direct effect on seed cotton yield. Hence selection for these traits would be quite effective to improve the seed cotton yield in upland cotton.

Dinakaran *et al.* (2012) conducted a study to assess the salt tolerance of 32 popular upland varieties released for general cultivation between 1980 and 2001 in India. The study was carried out in normal as well as saline-alkaline condition; in which salinity were created using bore well water irrigation and the average electrical conductivity level of bore well water is 3.10 ds/m. Correlation and path analysis studies revealed that the seed cotton yield was highly influenced by lint yield per plant in both normal and saline alkaline condition. Significant positive correlations exist between specific leaf area with leaf area index Bartlett's rate index with uniformity ratio, 2.5 per cent span length with bundle strength, uniformity ratio with micronaire and elongation percent. These results clearly indicated that selection for this trait might lead to concurrent improvement of other traits as well as seed cotton yield. The character's boll weight (-0.347), ginning out turn (-0.528), 2.5% span length (-0.312) and uniformity ratio (-0.440) registered high order negative direct effect on seed cotton yield. This result further confirmed the negative association between the quality and yield.

Song *et al.* (2012) evaluated inheritance and interrelationship of phenotype and genotype of earliness traits in a diallel analysis involving six early-maturing parents. Date of first

square (DFS), date of first flower (DFF), date of first open boll (DFOB), number of node first sympodial branch (NNFSB), and harvested rate before frost (HRBF) as earliness traits of six parents, 15 F₁ hybrids and 15 F₂ progenies were investigated from 2005 to 2008. The experiment design was a randomized complete block design with three replications. Correlation analysis showed generally that HRBF had a significant negative genetic and phenotypic correlation with DFS, DFOB, and NNFSB; DFS had significant positive genetic and phenotypic correlations with DFF, DFOB, and NNFSB; significant positive genetic and phenotypic correlations were also detected between DFF and DFOB, DFF and NNFSB, DFOB and NNFSB. The results showed that the lower the node to the first fruiting branch and the shorter the plant, the earlier was the onset of squaring, flowering, and boll opening, the higher was the harvest rate before frost.

Ahmad *et al.* (2008) examined 13 upland cotton genotypes to make rapid improvement in earliness, correlation between choice parameters related with earliness were determined. Highly positively significant correlation existed between days taken to first flower and sympodial branch node number with first effective boll ($r = 0.807$), days taken to first flower and boll opening percentage at 120 days after planting ($r = 0.705$), sympodial branch node number with first effective boll and boll opening percentage at 120 days after planting ($r = 0.501$), whereas negative correlation was found between sympodial branch length and boll opening percentage at 120 days after planting ($r = -0.663$), boll weight and seed cotton yield ($r = -0.281$).

2.8 Review of literature on multivariate analysis in upland cotton

Bilal *et al.* (2024a) opined that plant genetic diversity aids in the creation of new varieties that are more resistant to pests and unfavorable climatic conditions. In order to discover varied parents and assess their cross performance, the study was carried out. Principal component analysis (PCA) showed that PC-I and PC II accounted for 66.6% of the total variance. Plant height and green bolls were found to be positively correlated by PCA biplot analysis, but main stem nodes and open bolls showed a negative link, particularly with cotton leaf curl virus diseases. Based on cluster analysis, genotypes were divided into four clusters. Cluster-IV stood out because to its strong resistance to the cotton leaf curl virus disease and high yield, making it a good candidate for further breeding efforts. Other clusters include Mac-07, MNH-875, CRP-257, Super Okra, MNH-Super Gold, and GT/Bt Okra.

Bilal *et al.* (2024b) assessed the genetic diversity of 61 crosses in terms of yield and yield-related traits. Genetic diversity among germplasm forms the foundation of any breeding program and is pivotal for increasing yield. Principal component analysis revealed a total variance of 53% among crosses, with PC-I contributing 34.3% and PC-II contributing 18.7% of yield-related traits. Of eight PCs the four PCs exhibit > 1 eigen values. The number of squares per plant, the number of green bolls, and the number of flowers had positive effects on the first two PC components. Clusters I and II exhibited maximum values for the number of squares, the number of green bolls, the number of flowers, and resistance to cotton leaf curl virus disease. Crosses in cluster II displayed reasonable values for the number of open bolls and the number of flowers but showed susceptibility to cotton leaf curl virus disease. The crosses like, IS-29 $\times$ VR-5, 217007 $\times$ VR-5, IS-12 $\times$ VR-5, (MNH-1090 $\times$ MNH-875) $\times$ 875, (MNH-1090 $\times$ MNH-875/4) $\times$ 875/4, (MNH-872 $\times$ SS 32) $\times$ 872, CRP-255 $\times$ Super Gold, CRP-212 $\times$ FH-333, (MNH-872 $\times$ SS-102) $\times$ 872, (MNH-872 $\times$ MNH-875/4) $\times$ 875/4, CRI-288 $\times$ CRI-294, and CRI-287 $\times$ CRI-212 were genetically diverse for most of the studied morphological traits.

Çakmak *et al.* (2024) performed PCA analysis which indicated that fiber fineness and ginning out-turn were on the same platform, whereas seed cotton yield per plant with fiber length and fiber fineness were on opposite platforms.

Chapara *et al.* (2024) executed a study during Kharif, 2022 to evaluate the genetic variability of yield and its associated traits in cotton. The first three principal components (PCs) out of the 12 PCs displayed eigen values more than one and exhibited maximum share to total variability. The attributes that contributed a maximum portion to the total divergence included the number of sympodia per plant, lint yield, seed cotton yield, ginning outturn percent, and number of bolls per plant. The traits, number of bolls per plant, number of sympodia per plant, seed index, ginning out turn, and lint yield can be utilized for direct selection for further heterosis breeding. Based on D^2 analysis the lines, L 2251 and L 2252 were found divergent and might be used in further heterosis breeding programs.

Vadodaria *et al.* (2024) conducted a study comprising of 30 *G. arboreum* accessions to estimate the genetic variability using morphological traits. 30 genotypes were classified into 3 classes based on the seed cotton yield, which is considered a very important trait.

Manhattan dissimilarity co-efficient based phenotypic diversity divided the 30 genotypes into four main clusters, with an average dissimilarity value of 0.17, indicating low phenotypic variability.

Vishnuvardhan *et al.* (2023) evaluated 17 cotton genotypes with a view to study of genetic parameters for different yield and yield parameters. The results of principal component analysis revealed that, 4 Principal Components (PCs) were established with Eigen value greater than 1.00 which accounted for 83.9 % of the total variation for discriminating the lines. From principal component analysis, PC1 showed highest amount of variance (33.1%) with mostly related to traits like boll weight, seed index, lint index and halo length indicated the importance of these traits in relation to yield enhancement. Cluster analysis classified the genotypes into five clusters among which cluster I was largest with eight genotypes followed by cluster III and cluster IV with four and three genotypes respectively indicating the versatility of the genotypes of these clusters in the exploitation of heterosis.

Abdel-Monaem *et al.* (2022) undertaken a study in cotton (*G. barbadense* L.) to detect the magnitude of genetic divergence among parental cotton genotypes and their F₁s cross combinations and to select most suitable combinations for tolerant water deficient. The study was carried out at Sakha Agricultural Research Station, Kafr El-Sheikh, Agric. Res. Center, Egypt, during 2014 and 2015 growing seasons. The data showed that the first eight main principal components with eigen value more than one are extracted from the complicated components, the total cumulative variance of these eight factors amount for about 85.28% of the total multivariate variance. The female parent 10,229 formed wide cluster having wide divergent distance from the other groups. On the same trend, the extra-long varieties Giza 96 and Giza 45 formed unique groups, cluster number 8 and cluster 1 nearly related. Cluster number 7 and cluster 4 which consisted of two genotypes for each characterized by susceptible to water deficit conditions. The cross combinations Giza 77 x Dandra, Giza 77 x Pima S6, Giza 96 x Australy and Minufy x Dandra grouped at the same cluster, however the cross combinations Giza 86 x Dandra and Giza 68 x 10,229 classified into two different clusters. The data revealed that the inter cluster distance was higher than the intra cluster, indicating wide genetic divergent among the studied genotypes.

Balarabe *et al.* (2022) evaluated one hundred cotton genotypes from diverse sources for genetic improvement of important agronomic and yield traits in cotton. Hence, the need to study and explore available germplasm resources to assess the variation among the cotton germplasm and to classify and identify the potential genotypes that can be used in the development of hybrid with the best agronomic and yield traits. The experiment was conducted in a 10 x 10 lattice design, replicated twice across two environments in 2020 wet season. Principal component analysis (PCA) indicated that four components explained over 99.5% of the total variation of the data, with PC1 and PC2 accounting for 86.5% and 10% of the total variability, respectively. PC1 was associated with most of the traits except stand count at emergence, whereas PC 2 was related to stand count at emergence. Cluster analysis produced three clusters, cluster 1 consisted of genotypes (n = 39) that had low values for most of the traits assessed. Cluster 2 was composed of genotypes (n = 35) with high values of traits measured while cluster 3 had genotypes (n = 26) with the highest values of measured traits.

Seyed *et al.* (2022a) conducted a study in the Moghan Agricultural Research Station affiliated with the Ardabil Agricultural and Natural Resources Research and Training Center, Iran. In this study, 35 cultivars of cotton were evaluated for fiber-and yield-related traits during two consecutive years. Based on the ward's cluster analysis, the cotton cultivars were grouped into three different clusters. The cultivars in cluster 3 including Nazilli, No.228, No.200, B557, Armaghan, Golestan, and Shayan are superior in terms of their number of the boll, boll weight, earliness percentage, fiber percentage, fiber length, fiber fineness, fiber strength, fiber traction, and yield performance.

Seyyed *et al.* (2022b) evaluated 35 cultivars of cotton for fiber-and yield-related traits during two consecutive years. Based on the ward's cluster analysis, the cotton cultivars were grouped into three different clusters. The cultivars in Cluster 3 including Nazilli, No.228, No.200, B557, Armaghan, Golestan, and Shayan are superior in terms of their number of the boll, boll weight, earliness percentage, fiber percentage, fiber length, fiber fineness, fiber strength, fiber traction, and yield performance.

Meena *et al.* (2022b) characterized the genetic diversity of twenty-five genotypes of cotton and the genotypes were evaluated in randomized complete block design with four replications during kharif, 2019 at Agronomy Instructional Farm, SDAU, Sardarkrushinagar. The results of multivariate analysis revealed that the genotypes

grown had significant genetic divergence and the genotypes grouped into six clusters using Tocher's method in D^2 analysis. The cluster with the maximum genotypes was cluster I, followed by cluster VI. Cluster VI had the greatest intra-cluster distance and maximum inter cluster distance was calculated between cluster V and cluster VI. Inter cluster distances were greater than intra cluster distances, indicating that the genotypes differed significantly. Crosses between genotypes of these clusters can be made based on these studies to create suitable transgressive segregants. According to Mahalanobis' D^2 statistic, lint yield per plant and number of bolls per plant contributed the most to the overall genetic divergence.

Nadeem *et al.* (2022) investigated twenty exotic accessions of cotton to check their variability for within-boll yield components. The experiment was performed at the research area of the department of Plant Breeding and Genetics, University of Agriculture, Faisalabad. The genotypes were planted in three replications following a Randomized Complete Block Design (RCBD). At maturity, data were collected for different within boll yield parameters. Principal component analysis revealed that genotype ZB-18015 showed maximum variation for all parameters because it was at a maximum distance from the origin, followed by genotypes ZB-18020, ZB-18019 and ZB-18018, respectively. Cluster analysis identified particular groups of genotypes that had maximum similarity with each other.

Saraswat *et al.* (2022) characterized 50 elite lines of cotton during kharif 2021 at CCSHAU, Kaul for better documentation and utilization in breeding programmes. Biplot analysis observed 40.3% variation accounted by first two components with 22.5 %, 17.8% contributions. Cotton seed yield, number of bolls per plant, lint index, seed index, number of monopods contributed more in first principal component whereas ginning, locules per boll, lint index, number of monopods, plant height contributed more to second one.

Sahar *et al.* (2021) performed a study to assess the genetic diversity of yield and fiber quality traits in cotton. Analysis of variance exhibited significant variation for all studied traits except total number of nodes and the height to node ratio. The first 6 principal component analysis (PCs) out of the total fourteen PCs displayed eigenvalues (> 1) and had maximum share to total variability (82.79%). The attributes that had maximum share to total divergence included plant height, uniformity index, the number of sympodial

branches, seed per boll, GOT, seed cotton yield, and short fiber index. The genotype AA-802, IUB-13, FH-159, FH-458, and CIM-595 were genetically diverse for most of the yield and fiber quality traits and could be utilized for the selection of better performing genotypes for further improvement.

Sarwar *et al.* (2021a) carried out a study at Cotton Research Station, Ayub Agricultural Research Institute, Faisalabad, Pakistan during 2019 to estimate the genetic variation in 6 genotypes of cotton at three different sowing dates regarding cotton quality and yield traits. In principal component (PC) analysis out of 13 PCs, first three contributed 69.4(%) towards variability. PC I was important as yield had maximum negative factor loading. Further, there was important contribution of plant height, bolls count and sympodial branches count in PC I.

Sarwar *et al.* (2021b) performed principal component analysis (PCA) and cluster analyses to estimate genetic diversity in 25 cotton genotypes. In principal component analysis, four out of 14 principal components were selected with Eigen value >1. The four principal components contributed 79.9% towards variability. In PC-I, there was maximum positive factor loading of earliness traits, fiber fineness and negative factor loading of sympodia per plant and seed cotton yield. In PC-II, there was maximum positive factor loading of bolls per plant and GOT % while negative factor loading for fiber length. A biplot between PC-I and PC-II showed that major contribution towards variability of fiber fineness and earliness traits among the studied genotypes. In cluster analysis, 25 genotypes were allocated in four clusters. Cluster-I was the largest by having 10 genotypes while clusters-III and IV each having six genotypes, and cluster-II was the smallest having three genotypes

Sarwar *et al.* (2020) examined thirty-three genotypes from F₄ were assessed for earliness, high yield and good quality of fiber at Cotton Research Station, AARI, Faisalabad, Pakistan during 2018-19. For evaluation of these genotypes Principal Component analysis (PCA) and cluster analysis were performed to select suitable genetic material to utilize them in further breeding programs. The Principal Component Analysis showed first six Principal components having eigen value >1 showing 81.6 % of total variation with bolls count, weight of bolls, sympodial branches count and yield as being most related to PC I. Cluster analysis divided 33 genotypes into four divergent groups. Cluster 3 showed good values for bolls characters, fiber quality and yield and cluster 4 showed

good values for earliness. The genotypes in cluster 2, 3 and 4 could be combined to get desired traits combination.

Ullah *et al.* (2020) investigated one hundred and two germplasm of upland cotton for genetic divergence regarding yield related attributes using principal component analysis. The research was carried out in RCB design with 2 replications. Experiment data was recorded on various qualitative and quantitative parameters and were subjected to principal components analysis (PCA) and cluster analysis. Results: PCA result showed that only four components were considered on account of their eigenvalue greater than 1 which contributed 65% to the total variability. Score plot showed that the suncrop-6, tipu-9, TJ-max, Deebal, CRIS-543, TH20, Tahafuz-7, Eagle, BS-80, IUB-69, BH-221, NIAB-1048, and NIAB BT-2 showed the vertex of polygon and resulted as most divergent germplasm. Similarly cluster analysis also categorized the yield related traits into 5 main cluster. Cluster-1 contains 20 germplasm, cluster-II contain 16, and cluster-III, cluster-IV, and cluster-V comprise 13, 16, and 37 germplasm, respectively.

Bhatti *et al.* (2020b) evaluated fifteen upland cotton genotypes for their genetic potential under semi-arid conditions through two different multivariate analyses i.e. Principal component analysis (PCA) and Agglomerative hierarchical clustering (AHC) to characterize cotton genotypes on the basis of their performance. First four PCs contributed eighty-five (85%) variation and hence emphasized the role of plant height (PH), days to first flower (DFF), sympodial branches per plant (SBP) and monopodial branches per plant (MBP) for maximum variation. In PC1 genotypes possess maximum yield relating traits, LCA revealed two groups of genotypes, findings of PCA agreed with that of LCA. Information obtained through correlation, PCA and LCA would be used to devise breeding strategy for development of genotypes having better yield and lint quality.

Akter *et al.* (2019) carried out an experiment at the experimental field of Cotton Research, Training and Seed Multiplication Farm, Sreepur, Gazipur during the cropping season of 2015–2016 with 100 genotypes to evaluate genetic diversity of cotton genotypes for short duration using field performance. The genotypes under study were grouped into ten clusters through multivariate analysis using GENSTAT-5. Cluster III contained maximum number of genotypes (16) while cluster X contained the least number of genotypes (7). The inter cluster distances were larger than intra cluster

distances in all cases suggesting wider genetic diversity among the genotypes of different clusters. The maximum and minimum inter cluster distances were observed between clusters II and V (10.78) and clusters VIII and IX (3.30), respectively. The results indicated diverse and close relationship among the genotypes of those clusters. Earliness index, single boll weight and days to boll opening showed the higher contribution to the genetic divergence among 19 characters. Based on the results of genetic diversity and earliness index, the genotypes from cluster II could be used as parent in hybridization program for the development of short duration cotton variety.

Rathinavel K. (2019) characterized 320 cotton (*G. hirsutum*) germplasm accessions for distinctiveness. Under field and laboratory, 26 qualitative traits and 14 quantitative traits were recorded. In the principal component analysis, five principal components (PCs) extracted had Eigen value > 1 and contributed 76.80% of variations among the cotton germplasm. The clustering using UPGMA showed 12 distinct clusters. Based on these, an accession of a particular group or clusters might be selected for exploitation of its yield potential and fibre quality.

Isong *et al.* (2017) studied the relationship of yield components conducted on sixty hybrids obtained from a cross of ten *G. hirsutum* and six *G. barbadense* in a Line x Tester fashion. Six Principal components showed Eigen value of more than one with an accumulated variability of 70.39%. The Principal component with the highest variability was mostly related to seed cotton yield, uniformity ratio, boll weight, number of sympodia, number of bolls/plant and plant height.

Naik *et al.* (2016) studied genetic divergence among 50 genotypes of American cotton (*Gossypium hirsutum* L.) using Mahalanobis D^2 statistic and principal component analysis. On the basis of D^2 statistic, 50 genotypes were grouped into 8 clusters and PCA identified six principal components which explained 81.99% of the total variability among the genotypes. Based on intra and inter-cluster distance among the groups, it is suggested to make crosses between the genotypes of cluster VI (ARBH 1402) and cluster VII (HYPS 152), cluster I (SSGR 105) and cluster VIII (L 788), cluster I (SSGR 105) and cluster IV (L 799) and between cluster V (GJHV 497) and cluster VIII (L 788) for heterosis breeding. Divergence studies indicated that geographical diversity is always not necessarily associated with the genetic diversity. Multivariate analysis revealed the

presence of maximum divergence among L 788, HS 294, MCU 5, HYPS 152 and GISV 267 reporting their parental eligibility in exploitation of hybrid vigour.

Liaqat *et al.* (2015) investigated seventy-five genotypes of cultivated cotton (*Gossypium hirsutum* L.) for morphological characteristics i.e plant height, monopodial branches, sympodial branches, boll weight, seed volume, seed density and seed index. The principle component analysis (PCA) revealed significant differences between genotypes and the first four components with Eigen values greater than 1 contributed 66.68% of the variability among the genotypes.

Pujer *et al.* (2015) studied genetic divergence among 68 genotypes of American cotton (*Gossypium hirsutum* L.) using D^2 technique which indicated the presence of substantial genetic diversity among the material. The 68 genotypes were grouped into 8 clusters, based on the inter cluster distances C I and C VI were found to be most divergent followed by C V and C VI, C I and C VIII and C II and C VI. The use of diverse genotypes included in these clusters is advocated in crossing programme to evolve high heterotic hybrids and for taping the good transgressive segregants in segregating generations.

Reddy *et al.* (2015b) conducted an experiment with 63 genotypes of cotton to assess the genetic diversity for seventeen characters at Regional Agricultural Research Station, Lam Farm, Guntur, Andhra Pradesh. The 63 genotypes were grouped into eight clusters based on hierarchial cluster analysis. Among the clusters, cluster I was the largest with 13 genotypes followed by cluster VI with 11 genotypes. Cluster VII had minimum intra cluster Euclidean 2 distance value while, the inter cluster Euclidean 2 distance was highest between the clusters VI and VIII indicating the usefulness of the genotypes of these clusters in the exploitation of heterosis. In principal component analysis, first seven principal components with eigen value more than one contributed 84.00% towards the total variability. PC1 contributed maximum towards the total variability (23.799).

Sundar *et al.* (2014) evaluated one hundred and fifty-six cotton genotypes representing different plant types were studied for genetic diversity on the basis of quantitative and qualitative traits using Mahalanobis D^2 statistic. Out of ten clusters obtained, cluster I was the largest with fifty-nine genotypes, cluster II had fifty-five genotypes, cluster III had ten genotypes, cluster V had 13 genotypes, cluster VI had twelve genotypes, cluster VII had three genotypes while the remaining four clusters had single genotype each.

Cluster X exhibited the maximum mean values for seed cotton yield, number of bolls per plant and number of sympodia per plant.

Asha *et al.* (2013) conducted an experiment to analyze the genetic diversity among 40 genotypes for 15 quantitative characters which were grouped into 7 clusters based on Mahalanobis D^2 analysis with cluster I and III being the largest each with 8 genotypes followed by 7 genotypes in cluster VII. Based on Hierarchical cluster analysis all the genotypes were grouped into 7 clusters with cluster VII being the largest each with 13 genotypes followed by 8 genotypes in cluster IV. This random distribution of genotypes indicated absence of parallelism between geographical and genetic diversity. In Principal component analysis first seven principal components with eigen value more than one contributed 87.98 per cent towards the total variability with PC1 alone showing maximum of 22.27 per cent variability.

2.9 Review of literature on molecular profiling of upland cotton

Shinde *et al.* (2025) selected a total 25 cotton accessions and 25 SSR markers to depict diversity and correlation between the genotypes for fiber quality traits. The study was conducted at Biotechnology Laboratory College of Agriculture, Pune and Central laboratory of ICAR-Directorate of Floricultural Research, Keshavnagar, Pune. Out of 25 SSR markers, 17 were amplified and all the markers were polymorphic. Total 71 loci amplified by 17 markers. Out of these, 59 were polymorphic and 12 were monomorphic. The polymorphic information content (PIC) value ranged from 0.18 to 0.82, maximum PIC recorded for JESPR-289 and minimum for CIR-089. The similarity matrix and dendrogram was constructed using Jaccard's similarity coefficient with NTSYS 2.02i software. The value of coefficient of similarity varied from 0.46 to 0.97. The dendrogram revealed that two major clusters were formed among 25 cotton genotypes. Cluster I contain twenty-one genotypes while Cluster II contain four genotypes. The correlation was calculated. Total of 7 markers were found to be correlated with fiber quality. Out of that seven markers, BNL-3090 and BNL-3867 were found to positively correlated with all three fiber quality parameters. The remaining five BNL-1059, BNL-3147, BNL-3435, BNL-3510 and CIR-413 were positively correlated with length and strength. BNL-3090 and BNL-3867 are only markers found to be correlated with fineness.

Altaf *et al.* (2024) investigated 28 EST-SSR markers to unveil the genetic diversity of 30 upland cotton genotypes collected from Central Cotton Research Institute, Multan, Pakistan. Seven EST-SSR primers out of 28 were found polymorphic at different SSR loci. A total of 37 alleles were ascertained using the 28 EST-SSR primers. The ordinary number of alleles per locus was 1.5, with a range from 1 to 3. The polymorphic information content (PIC) extended from 0.065 to 0.82 with an average of 0.403. Cluster analysis was executed using an un-weighted pair group method with arithmetic. The average algorithm grouped cotton genotypes into four major clusters except DNH-105, Cyto-178 and FH-326, acting as an out-group. Genotypes such as IR-3701, Sitar-008, CIM-598, and CIM-625 exhibited maximum similarity coefficient. Nau-1231 indicated the lowest confusion probability (Cj) value (0.531) along with the greatest PIC (0.82) and discriminating power (Dj) (0.82) values.

Revanasiddayya *et al.* (2024) employed 50 simple sequence repeat (SSR) markers linked to the yield and fibre quality/colour traits for estimating the genetic diversity in 33 cotton genotypes of diploid and tetraploid species differing in fibre colour. The diversity analysis was performed in GenAlEx (Ver. 6.41) and Powermarker (Ver. 3.25) while DARwin (Ver. 6.0.21) software was used to establish the phylogenetic relationships following neighbour-joining (NJ) and unweighted pair group method with arithmetic (UPGMA) mean method. Markers generated 186 polymorphic loci as genotypic data with an average of 3.72 alleles and an average polymorphic information content (PIC) value of 0.59 per SSR locus. The NJ and UPGMA grouped 33 genotypes into three major clusters I, II and III consisting of 21 tetraploid *Gossypium hirsutum*, 10 *G. arboreum* coloured and 2 white cotton genotypes, respectively. In the PCA, the first two components (PC1 and PC2) explained 74.69% of the variation and the biplot plotted the 33 genotypes in three groups. The study established the diverse nature of 33 cotton genotypes based on their fibre colour and ploidy level.

Vadodaria *et al.* (2024) conducted a study for molecular diversity analysis using 24 SSR markers carried out for the 30 *G. arboreum* genotypes and 11 additional cotton species. The dendrogram generated using SSR marker data based on Nei's genetic distance grouped 41 genotypes into three clusters, with average distance of 0.47. Based upon the placement of the number of genotypes in same clusters in both matrices, it could be inferred that there was considerable correlation between morphological and molecular analysis as the value for Mantel Statistic R was 0.42 (p value 0.0003).

Aydin & Gucer (2023) used high-resolution capillary gel electrophoresis to determine the genetic diversity of *G. hirsutum* and *G. barbadense* genotypes. Using 19 EST-SSR markers, a total of 47 genotypes were screened. The PIC values of the markers used ranged from 0.268 to 0.889. The mean PIC value was calculated to be 0.603. Concerning the clustering, PCoA and population structure analyses gave similar results, and the genotypes could be divided into 3 main groups. Genetic admixture with *G. hirsutum* was found in some genotypes of *G. barbadense* species.

Iqbal *et al.* (2023) compared the effectiveness of EST-SSRs and ISSRs markers for assessing genetic variation among 45 Pakistani Bt and non-Bt cotton varieties. ISSR and EST-SSR primers yielded 108 and 28 loci, respectively. The polymorphism was found 32.40% for ISSR, while 89.28% was recorded for SSR primers. Cluster analysis revealed a high level of genetic similarity for ISSR (average 0.92) and EST-SSR (average 0.85) among cotton genotypes. The mean polymorphic information content (PIC) value was 0.25 for ISSR, whereas it was recorded 0.48 for EST-SSRs. Confusion probability (C_j) exhibited a negative association with discriminating power (D_j), while D_j displayed a positive association with PIC. Marker discriminating statistics showed that EST-SSRs have a high expected heterozygosity of polymorphic loci (H_{ep}) as compared to ISSR, along with a higher marker index value (MI). The effective multiplex ratio for ISSRs (1.40) was greater than EST-SSRs (1.12). The structural analysis revealed 6 sub-clusters for EST-SSRs and 4 sub-clusters for ISSRs. This phylogenetic study is crucial for identifying promising genotypes for breeding programs, especially given the limited genetic diversity in cotton breeding. The study showed that Bt cotton genotypes share a high genetic similarity, emphasizing the need for introducing diverse or exotic genotypes into breeding programs to enrich genetic diversity. Additionally, marker-discriminating indices could aid in selecting effective markers to assess genetic variation, facilitating the development of improved cotton varieties with desired traits.

Shi *et al.* (2023) carried out a molecular marker research on 191 upland cotton germplasm resources by using 26 simple sequence repeat (SSR) markers to analyse polymorphism and population structure. A total of 92 clearly identifiable bands were amplified from 65 SSR markers, with an average of 3.54 per marker. The mean number of observed alleles (N_a) was 1.984, the effective allele number (N_e) was 1.534, the gene diversity index (h) was 0.312, the Shannon–Weaver index (I) was 0.470, and the polymorphism information content (PIC) of the primers was 0.530. Cluster analysis

divided the germplasm into 11 groups at a genetic distance of 0.52, and group 1 was divided into 6 subgroups at a genetic distance of 0.39. The germplasms from different regions were interspersed among the various groups, indicating that the germplasm resources within cotton regions were also quite different. All materials were divided into three populations by structure analysis, and there was gene exchange between the three groups. The above results showed that the DNA clustering of upland cotton germplasm was not related to its geographical origin but to its genetic relationships, thus indicating the genetic diversity and complexity of cotton germplasm.

Çelik. (2022) evaluated some of Upland cotton varieties which were screened with 20 polymorphic SSR primers, and their population structure and genetic diversity analysis were examined. 17 SSR primers amplified 99 alleles with a 5.82 allele per locus. The mean PIC value of the markers was 0.312. The highest PIC value (0.491) belongs to the Nau3736 SSR marker while Bnl1611 and Bnl3449 markers had the lowest PIC value (0.105). The Genetic Distance (GD) values of the markers varied between 0.26 and 1.09. The highest GD values were between Sure Grow 96 and Carmen, Sealand-542 and Siokra ¼, and between Sphinx V and Stoneville-453 cultivars. As a result, the genetically distant cultivars (Acala maxx, Carmen, Aleppo 40, Siokra ¼, and Tex) might be recommended to use as parents in Marker-assisted selection (MAS) technology to develop new cotton varieties which were resistant or tolerant to stress factors.

Kuang *et al.* (2022) performed DNA fingerprinting and genetic diversity analysis of 79 early-maturing upland cotton (*Gossypium hirsutum* L.) cultivars using Simple Sequence Repeat (SSR) molecular markers. Out of 126 pairs of SSR primers, they selected 71 pairs that gave good polymorphisms and clear bands, had good stability, and showed even distribution on the cotton chromosomes, and 142 polymorphic genotypes were amplified. The average number of alleles amplified with the SSR primers was 2.01. The polymorphism information content (PIC) of the markers ranged from 0.1841 to 0.9043, with an average of 0.6494. The results of fingerprint analysis showed that nine varieties had characteristic bands, and at least six primer pairs could be used to completely distinguish all 79 cotton accessions. Using NTSYS-pc 2.11 cluster analysis, the genetic similarity coefficients between the cotton genotypes ranged from 0.3310 to 0.8705, with an average of 0.5861. All cotton accessions were grouped into five categories at a similarity coefficient of 0.57, which was consistent with the pedigree sources. At the same time, the average genetic similarity coefficients of early-maturing upland cotton

varieties in China showed a low-high-low pattern of variation over time, revealing the development history of early-maturing upland cotton varieties from the 1980s to the present. This also indirectly reflects that in recent years, China's cotton breeders have focused on innovation and have continuously broadened the genetic resources for early-maturing upland cotton.

Sarode *et al.* (2021) analyzed genetic diversity and relationships among 25 cotton (*Gossypium hirsutum* L.) genotypes using morphological and molecular markers. Morphological analysis was performed based on 3 fiber quality traits including fiber length, strength and fineness the mean values of each trait for each genotype were calculated. The results showed that most of the genotypes have long fiber length, very high fiber strength and average fiber fineness. Twenty-five SSR markers related with fiber quality traits were found to be polymorphic produced a total of 53 amplicons with an average of 2.12 alleles per locus ranging from 130 bp to 370 bp products, Markers showing 100% polymorphism with average polymorphism information content (PIC) value of 0.290. The BNL-1059 (PIC-0.481), CIR-244 (PIC-0.538), CIR-354 (PIC-0.660), CIR-381 (PIC-0.784), CIR-246 (PIC-0.749) and JESPR153 (PIC-0.639) marker were found to be most informative which indicates significantly diversity between all genotypes for fiber quality traits. Similarity matrix was used to depict dendrogram through UPGMA cluster analysis based on SSR similarity coefficient ranged from 0.61 to 0.91 indicating the fact that large proportions of genotype were dissimilar. The present study revealed that genotypes BN-1, PH-348, and ARB-908 for fiber quality were found to be the most diverse having a broad genetic base for the concerned fiber traits.

Modi *et al.* (2020) carried out a study to determine genetic variability through DNA markers in 26 cotton germplasm. The genetic variability and relationships among 26 cotton germplasm were investigated using SSR markers. 17 SSR primers generated a total 36 alleles, of which 30 alleles were found to be polymorphic, resulting 83.33% polymorphism. The similarity coefficient based on SSR markers ranged from 0.36 to 0.92 with an average of 0.43, thus suggesting considerable genetic variation between the cotton germplasm studied. Dendrogram was generated by UPGMA cluster analysis based on Jaccard's similarity coefficient, grouped 26 germplasm lines into two distinct clusters comprising of cluster A and cluster B.

Shoukat *et al.* (2020) studied genetic diversity in representative sets of advanced cotton genotypes at molecular level employing polymorphic simple sequence repeats. Of seventy-two advanced cotton genotypes, thirty microsatellites primer pairs were studied, 29 showed to be polymorphic. DNA markers amplified fragments of 100 to 300 bp in size. The polymorphism information contents ranged between 0.76 and 0.99. Cluster analysis clearly grouped the 72 genotypes into seven groups A, B, C, D, E, F, G based on Nei's coefficient similarity matrix. Group B was found to be the most genetically distinct group when compared to other groups. However, group B had most of the candidates of under observed genotypes. This study exposed two major clusters of cotton cultivars and recognized the cotton genotypes which were genetically different.

Wu *et al.* (2020) aimed to develop a set of perfect simple sequence repeat (SSR) markers with a single copy in the cotton genome, to construct a DNA fingerprint database suitable for authentication of cotton cultivars. They optimized the polymerase chain reaction (PCR) system for multi-platform compatibility and improving detection efficiency. Based on the reference genome of upland cotton and 10× resequencing data of 48 basic cotton germplasm lines, single-copy polymorphic SSR sites were identified and developed as diploidization SSR markers. The SSR markers were detected by denaturing polyacrylamide gel electrophoresis (PAGE) for initial screening, then fluorescence capillary electrophoresis for secondary screening. The final perfect SSR markers were evaluated and verified using 210 lines from different sources among Chinese cotton regional trials. Using bioinformatics techniques, 1 246 SSR markers were designed from 26 626 single-copy SSR loci. Adopting a stepwise (primary and secondary) screening strategy, a set of 60 perfect SSR markers was selected with high amplification efficiency and stability, easy interpretation of peak type, multiple allelic variations, high polymorphism information content (PIC) value, uniform chromosome distribution, and single-copy characteristics. A multiplex PCR system was established with ten SSR markers using capillary electrophoresis detection. A set of perfect SSR markers of cotton was developed and a high-throughput SSR marker detection system was established. This study laid a foundation for large-scale and standardized construction of a cotton DNA fingerprint database for authentication of cotton varieties.

Yuzhen *et al.* (2020) aimed to develop a set of perfect simple sequence repeat (SSR) markers with a single copy in the cotton genome, to construct a DNA fingerprint database suitable for authentication of cotton cultivars. They optimized the polymerase

chain reaction (PCR) system for multi-platform compatibility and improving detection efficiency. Based on the reference genome of upland cotton and 10× resequencing data of 48 basic cotton germplasm lines, single-copy polymorphic SSR sites were identified and developed as diploidization SSR markers. The SSR markers were detected by denaturing polyacrylamide gel electrophoresis (PAGE) for initial screening, then fluorescence capillary electrophoresis for secondary screening. The final perfect SSR markers were evaluated and verified using 210 lines from different sources among Chinese cotton regional trials. Using bioinformatics techniques, 1 246 SSR markers were designed from 26 626 single-copy SSR loci. Adopting a stepwise (primary and secondary) screening strategy, a set of 60 perfect SSR markers was selected with high amplification efficiency and stability, easy interpretation of peak type, multiple allelic variations, high polymorphism information content (PIC) value, uniform chromosome distribution, and single-copy characteristics. A multiplex PCR system was established with ten SSR markers using capillary electrophoresis detection. A set of perfect SSR markers of cotton was developed and a high-throughput SSR marker detection system was established.

Manonmani *et al.* (2019) studied the molecular diversity of twelve cotton genotypes, in order to select the suitable divergent parent for heterosis breeding. Totally, 55 primers were used to diversify these genotypes and 40 pairs showed clear, scorable and unambiguous bands. Out of 40, 25 primer pairs (62.55 %) were found to be polymorphic and 15 found to be monomorphic. The average number of alleles per locus was 1.8. The similarity coefficient ranged from 0.56 to 1.00 with high dissimilarity coefficient of 0.44 for the genotypes TCH 1777 with MCU 13 and TSH 0499 with SVPR 4. A high similarity coefficient of 0.92 was observed among the parents BGDS 1063 with ARBC 19 and TSH 0499 with TSH 1819. The results of cluster analysis grouped twelve genotypes into seven clusters. The largest cluster was cluster A, which had four genotypes and cluster B had two genotypes. The cluster C, D, E, F and G had one genotype type each.

Santhy *et al.* (2019) assessed genetic diversity in 48 popular varieties of tetraploid cotton from each cultivated zone of India using 68 SSR markers distributed across linkage groups. The markers produced a total of 144 alleles with an average of 2.19 per locus. The polymorphism information content (PIC) ranged from 0.04 to 0.57 with a mean of 0.23 indicating lesser diversity in the studied material. Jaccard's similarity index based

neighbourhood joining cluster analysis grouped the genotypes into three major clusters, each of which was further classified into sub-groups. Inconsistencies were observed between the clusters and known pedigree of the cultivars. A narrow genetic base was also revealed among the cotton cultivars. The SSR markers revealed a genetic similarity of 73% among the varieties studied. The DNA fingerprint developed using a selected set of 14 markers showed a probability of identical match of 2.47×10^{-3} with high goodness of fit ($r^2 = 0.86$).

Li *et al.* (2018) selected 26 out of 83 simple sequence repeat markers associated/linked with cotton important agronomic traits derived from previous and other published research, corresponding to the 26 chromosomes of upland cotton (*Gossypium hirsutum* L.), as core primers for DNA fingerprinting construction. The 26 markers showed clear band patterns, good repeatability and high polymorphism. The average alleles, gene diversity index and polymorphism information content were 3.12, 0.4312 and 0.3830, respectively. Using TM-1, a genetic standard line for upland cotton, as the control, DNA fingerprinting pattern and DNA barcodes were obtained based on the core primers. There was a significant positive correlation between genetic distance matrix determined using 26 core primers and that determined using more primers (335) derived from previous research, further suggesting that the core primers were eminently suitable for DNA fingerprinting in upland cotton.

Fu *et al.* (2017) screened 5000 pairs of primers and chose 17 pairs of core simple sequence repeat (SSR) primers to determine the F_1 purity of Han6402. The results showed that the purity based on SSR markers reached 100%. Twelve of the 17 pairs of primers exhibited co-dominant banding patterns, and 5 showed non co-dominant banding patterns. Moreover, we constructed an F_1 SSR fingerprinting profile that enabled the identification of the authenticity of Han 6402. Using these primers, we subsequently detected 44 individual F_2 seedlings, and the results exhibited different extents of separation, in which the majority of genotypes were heterozygous with co-dominance at most of the loci that differed from each other. The results validated the underlying heterozygous status of the F_2 population at the molecular level. Therefore, they concluded that the set of core SSR primers could be used for the laboratory identification of the authenticity and purity of cotton hybrids, not only for distinguishing F_1 hybrids or segregating F_2 populations, but also for detecting volunteer seeds as fake F_1 hybrids in the cotton hybrid industry, based on the hybrid fingerprinting.

Pereira *et al.* (2015) initially genotyped, 92 SSR markers in 20 Brazilian cotton cultivars to identify polymorphic simple sequence repeat (SSR) markers for varietal identification of cotton and evaluation of the genetic distance among the varieties. Of this total, 38 loci were polymorphic, two of which were amplified by one primer pair; the mean number of alleles per locus was 2.2. The values of polymorphic information content (PIC) and discrimination power (DP) were, on average, 0.374 and 0.433, respectively. The mean genetic distance was 0.397 (minimum of 0.092 and maximum of 0.641). A panel of 96 varieties originating from different regions of the world was assessed by 21 polymorphic loci derived from 17 selected primer pairs. Among these varieties, the mean genetic distance was 0.387 (minimum of 0 and maximum of 0.786). The dendrograms generated by the unweighted pair group method with arithmetic average (UPGMA) did not reflect the regions of Brazil (20 genotypes) or around the world (96 genotypes), where the varieties or lines were selected. Bootstrap resampling shows that genotype identification is viable with 19 loci. The polymorphic markers evaluated were useful to perform varietal identification in a large panel of cotton varieties and might be applied in studies of the species diversity.

Qin *et al.* (2015) investigated 359 SSR (simple sequence repeat) markers for the association mapping of 241 Upland cotton collections. A total of 333 markers, representing 733 polymorphic loci, were detected. The average linkage disequilibrium (LD) decay distances were 8.58 cM ($r^2 > 0.1$) and 5.76 cM ($r^2 > 0.2$). 241 collections were arranged into two subgroups using STRUC TURE software. Mixed linear modeling (MLM) methods (with population structure (Q) and relative kinship matrix (K)) were applied to analyze four phenotypic datasets obtained from four environments (two different locations and two years). Forty-six markers associated with the number of bolls per plant (NB), boll weight (BW), lint percentage (LP), fiber length (FL), fiber strength (FS) and fiber micronaire value (FM) were repeatedly detected in at least two environments. Of 46 associated markers, 32 were identified as new association markers, and 14 had been previously reported in the literature.

Sundar *et al.* (2014) evaluated one hundred and fifty-six cotton genotypes representing different plant types were studied for genetic diversity on the basis of SSR marker analysis. Two distinct groups resulted from SSR marker data analysis. Among the 49 primers, BNVB-37421, CIR081, CIR081, CIR005, CIR182, CIR238, CIR413, JESPR29, NAU1278, NAU3561, NAU1262, NAU992, NAU1037 produced maximum number of

alleles. All the robust lines were clustered in cluster I (fifty-nine) and all the compact lines (fifty-five) were clustered in cluster II. The highest contribution towards genetic divergence was recorded by boll weight (gm), in 6917 cases out of 19506 combinations, accounting for 35.46 per cent followed by number of bolls per plant and seed cotton yield.

Tu *et al.* (2014) studied the genetic diversity of 51 upland cotton cultivars with different parental origins and breeding periods that were developed in Hubei Province on the basis of 237 mapped simple sequence repeat markers covering the cotton genome. A total of 108 polymorphic primer pairs amplified 196 loci; the polymorphism information content range was 0.04 to 0.83, with an average of 0.46. A model-based clustering analysis (STRUCTURE) of the genomic data identified 3 clear subpopulations, and the result was confirmed by principal components analysis. The genetic similarity coefficient among 51 upland cotton cultivars was 0.598 on average, ranging from 0.378 to 0.817. The unweighted pair group method with arithmetic average cluster analysis revealed inconsistencies in other clustering patterns: “Tianmian 1” was distinct from the rest of the materials and formed a separate cluster.

Ahmed *et al.* (2013) selected thirty-eight upland cotton cultivars and 55 hybrids from three cotton growing regions of China mainland i.e. yellow river cotton valley (YRCV), Yangtze river cotton valley (YzRCV) and north west dry region (NWDR); featured with perceptible climatology. Twelve randomly selected cultivar and hybrids (representative sample of the three regions) were employed to reveal polymorphism across mapped SSRs. Sixty-six genome-covered polymorphic SSRs were employed to assess genetic relatedness among all accessions. The results showed a polymorphism information content range from 0.34-0.86 and resolving power 0.04-2.45 for genome-covered SSRs. Higher PIC values rendered selection of 13 highly informative potential core SSRs, whose average PIC value was 0.80. Moreover, Jaccard's similarity coefficients of genome-covered and potential core SSRs were compared and found to be related. Potential core SSRs substantiated clustering results of genome-covered SSRs and successfully discriminated all accession. Overall clustering pattern and revealed genetic constitution of tested material suggested that the 13 potential core SSRs could assure comparable results with higher resolution as compared to that of genome-covered SSRs.

Ijaz *et al.* (2013) conducted a research to study the genetic relatedness among 20 cotton genotypes using simple sequence repeats markers using 31 simple sequence repeats (SSRs) belonging to BNL series. These 31 DNA markers amplified fragments of 80 to 340 bp in size through SSR profiling. Seven markers out of 31 were found to be polymorphic. A total of 41 loci were amplified out of these, 17 loci were informative showing 41% polymorphism. On average, 1.3 loci per primer were amplified. A dendrogram was constructed using pair group method of arithmetic means (UPGMA) method comprising eight main groups that is, A, B, C, D, E, F, G and H. Genotypes MNH-147 and GOHAR-87 emerged as genetically most similar with a value of 97% followed by 97% similarity between the genotypes CIM-443 and FVH 53. The genotypes CIM-1100 and BH-136 were found most divergent showing 58% genetic similarity.

Ullah *et al.* (2012) evaluated genetic divergence among 19 Bt cotton genotypes using simple sequence repeat (SSR) markers. Thirty-seven of 104 surveyed primers were found informative. Fifty-two primers selected on the basis of reported intra-*hirsutum* polymorphism in a cotton marker database showed a high degree of polymorphism, 56% compared to 13% for randomly selected primers. A total of 177 loci were amplified, with an average of 1.57 loci per primer, generating 38 markers. The amplicons ranged in size from 98 to 256 bp. The genetic similarities among the 19 genotypes ranged from 0.902 to 0.982, with an average of 0.947, revealing a lack of diversity. Similarities among genotypes from public sector organizations were higher than genotypes developed by private companies. Hybrids were found to be more distant compared to commercial cultivars and advanced breeding lines. Cluster analysis grouped the 19 Bt cotton genotypes into three major clusters and two independent entries. Cultivars IR-3701, Ali Akbar-802 and advanced breeding line VH259 grouped in subcluster B2, with very narrow genetic distances despite dissimilar parentage. We found a very high level of similarity among Pakistani-bred Bt cotton varieties, which means that genetically diverse recurrent parents should be included to enhance genetic diversity. The intra-*hirsutum* polymorphic SSRs were found to be highly informative for molecular genetic diversity studies in these cotton varieties.

Rakshit *et al.* (2010) screened a total 177 simple sequence repeat (SSR) markers using a set of 47 Upland cotton genotypes comprising 14 commercial varieties, 14 germplasm accessions and 19 advanced breeding lines to identify informative markers for genetic

diversity assessment and fingerprinting in *G. hirsutum*. Only 21% (38/177) of SSR markers tested showed polymorphism with a mean of 2.18 alleles per locus and with average polymorphism information content (PIC) of 0.32. The SSR markers revealed a Jaccard's similarity coefficient ranging between 0.43 and 0.89, with an average of 0.67 among accessions. Cluster analysis using unweighted pair group method with arithmetic averages (UPGMA) and principal component analysis (PCA) indicated that majority of the genotypes were very closely related. All the 47 genotypes showed heterozygosity for at least one of the SSR loci. We discovered 19 rare and 6 unique alleles among the tested genotypes of cotton. Fingerprint based on all the 38 loci revealed a probability of identical match by chance of 3.98×10^{-8} . A set of ten SSR markers was identified which could distinguish all the 47 genotypes with a moderate probability of identical match by chance ($X_D^n = 0.01$).

Hussein *et al.* (2007) investigated genetic variability and relationships among eleven cotton genotypes (8 *G. barbadense* and 3 *G. hirsutum*) using 28 SSR primers or primer combinations, which produced 117 amplicons. The number of polymorphic amplicons was 65. Therefore, the level of polymorphism among the eleven genotypes was 55.6. The genetic relationships among the 11 cotton genotypes was examined using the Dice coefficient to compute the similarity matrices. UPGMA analysis was performed and dendrograms were constructed. Based on the SSR data, the genetic similarities ranged from 13.8% to 98.1%. All dendrograms clustered the 11 genotypes into two main clusters: one containing the 8 genotypes belonging to *Gossypium barbadense* and the other one contained the 3 genotypes belonging to *Gossypium hirsutum*. Four genotypes were characterized by nine SSR markers. Furthermore, the different marker types permitted the discrimination between the two cotton species (*G. hirsutum* and *G. barbadense*) through revealing species-specific markers. A total of 25 SSR markers were specific to *G. hirsutum*. These species specific markers, might be related to the boll weight average and Pressly cotton characters.

CHAPTER III

MATERIALS AND METHODS

Cotton (*Gossypium hirsutum* L.) is a crucial raw material for textile industry. The available cotton varieties of Bangladesh are generally low yield potentials compared to world's top cotton producer countries due to narrow genetic base and lack of divergence in the existing cultivars. So, the aim of the experiment was to study the genetic divergence, variability, character association, fiber quality estimation and molecular characterization of upland cotton genotypes. To satisfy these goals, the experiments were sequentially completed according to the framework.

3.1 List of the experiments

Three experiments were conducted during the period from 2021 to 2023. The field trials were carried out in every cropping season in the speculated period of the investigations. The list of the experiments with their experimental tenure is presented in **Table 1**.

3.2 Morphological and fiber quality parameter estimation of upland cotton genotypes

3.2.1 Experimental tenure, location, climate and soil

The experiments were conducted from July-December in each year of 2021, 2022 and 2023. The field experiments were carried out in the experimental field of Cotton Research, Training and Seed Multiplication Farm, Sadarpur, Dinajpur (Appendix I). The quality parameters were estimated in the Head Office of Cotton Development Board (CDB), Dhaka. The geographical position of the field experiments was 25° 45' 19" North latitude and 88° 41' 32" East longitude, at an altitude of 34.5 meters above the average sea level (Appendix V). The experimental site was located in a subtropical climate characterized by average temperature (°C) and significant rainfall from July to September (Appendix II, III & IV). The area was under Tista Meander Floodplain Agro-Ecological Zone (AEZ-3) of Bangladesh and characterized by moderate fertile sandy loam soil with a pH of 5.86 (Appendix V).

Table 1. List of the experiments conducted in the tenure of 2021-2023

Exp.	Title of the experiment	Experimental materials	Season	Field	Laboratory
I	Characterization of 100 upland cotton (<i>Gossypium hirsutum</i> L.) genotypes to select fiber yield enhancing traits and promising early genotypes	100 upland cotton genotypes	July-December, 2021	√	-
II	Evaluation of 45 upland cotton (<i>Gossypium hirsutum</i> L.) genotypes for revealing early performances and measuring genetic diversity	45 upland cotton genotypes selected from 1 st year	July-December, 2022	√	-
III	Assessment of early boll picking, fiber quality and molecular profiling of well performing 12 upland cotton (<i>Gossypium hirsutum</i> L.) genotypes	12 upland cotton genotypes selected from 2 nd year	July-December, 2023	√	√

3.2.2 Experimental materials

A total of 100 presumed genetically diverged upland cotton (*Gossypium hirsutum* L.) genotypes collected from the gene bank of Cotton Development Board (CDB), Cotton Research Centre, Mahiganj, Rangpur, Bangladesh in 2021, were used in the first experiment (**Table 2**). The experimental materials comprising released varieties, elite breeding lines, popular cultivars, a mutant variety, and advanced lines selected based on their diverse morphological and agronomic performance. The second experiment was conducted in 2022, using the superior 45 lines selected from first year (**Table 3**). Then in 2023, the third experiment was conducted using the well performing 12 lines including two checks selected from second year (**Table 4**).

3.2.3 Experimental design and layout

The field experiments were laid out in a RCBD with three replications. 4 m × 4.5 m unit plot was planted with a spacing of 40 cm between plants and 90 cm between rows, with three seeds put in 10 lines for each genotype. The genotypes were randomly distributed in each of the plots (**Figure 2a**).

3.2.4 Field and other operations

3.2.4.1 Land preparation

The land was prepared by two ploughings and one cross ploughing with a tractor mounted disc plough. Three days later again the land was ploughed and cross ploughed with the country plough followed by laddering to get a good seed germinating conditions. Weed and stubbles were removed from the field prior to sowing of seeds. Manures and fertilizers were applied as per the recommended doses and irrigation channels were made around each block.

3.2.4.2 Seeds sowing

Experimental seeds were sown with three seeds put in 10 lines for each genotype in July 1 of each cropping season.

3.2.4.3 Intercultural operations

The following intercultural operations were carried out during entire cropping period for proper growth and development of the plants and to receive a good harvest.

Table 2. List of 100 upland cotton lines used in the first experiment in 2021

SL	Genotype	SL	Genotype	SL	Genotype	SL	Genotype
1.	RA-2	26.	CB-17	51.	JA-11/L	76.	BC-0038
2.	RA-4	27.	CB-1055	52.	JA-17/2	77.	BC-0039
3.	RA-5	28.	BC-0385	53.	JA-13/X	78.	BC-0040
4.	RA-9.N5	29.	BC-0397	54.	JA-09/G	79.	BC-0041
5.	RA-15	30.	BC-0410	55.	JA-08/4	80.	BC-0042
6.	RA-16	31.	BC-0415	56.	JA-085	81.	BC-0043
7.	SR-17	32.	BC-0419	57.	JA-0510	82.	BC-0044
8.	SR-18	33.	BC-0435	58.	Mutant-1 (Check)	83.	BC-0045
9.	SR-19	34.	BC-0436	59.	TC-1901	84.	BC-0046
10.	CB-1	35.	BC-0442	60.	TC-1902	85.	BC-0047
11.	CB-2	36.	BC-0462	61.	TC-1903	86.	BC-0048
12.	CB-3	37.	BC-0488	62.	TC-1904	87.	BC-0049
13.	CB-4	38.	BC-0490	63.	BC-0024	88.	BC-0050
14.	CB-5	39.	BC-0491	64.	BC-0025	89.	BC-0051
15.	CB-6	40.	BC-0495	65.	BC-0026	90.	BC-0052
16.	CB-7	41.	BC-0509	66.	BC-0027	91.	BC-0053
17.	CB-8	42.	BC-0510	67.	BC-0028	92.	BC-0054
18.	CB-9	43.	BC-0511	68.	BC-0029	93.	BC-0055
19.	CB-10	44.	BC-0512	69.	BC-0030	94.	BC-0056
20.	CB-11	45.	BC-0513	70.	BC-0031	95.	BC-0057
21.	CB-12	46.	BC-0514	71.	BC-0032	96.	BC-0058
22.	CB-13	47.	BC-0515	72.	BC-0033	97.	BC-0059
23.	CB-14 (Check)	48.	JA-11/N	73.	BC-0035	98.	BC-0060
24.	CB-15	49.	JA-13/R	74.	BC-0036	99.	BC-0061
25.	CB-16	50.	JA-08/B	75.	BC-0037	100.	BC-0062

Origin: Gene bank of Cotton Development Board (CDB), Cotton Research Centre, Mahiganj, Rangpur, Bangladesh.

3.2.4.3.1 Weeding

Weeding was done during first two side dressing of urea to break the soil crust, to keep the plots free from weeds and incorporate the urea properly into the soil which reduced the loss of urea through de-nitrification and leaching.

Table 3: List of the selected 45 lines used in second experiment in 2022

SL	Genotype	SL	Genotype	SL	Genotype
1.	RA-2	16.	Mutant-1 (Check)	31	BC-0046
2.	RA-5	17.	TC-1903	32	BC-0047
3.	RA-9.N5	18.	TC-1904	33	BC-0048
4.	CB-2	19.	BC-0024	34	BC-0049
5.	CB-6	20.	BC-0025	35	BC-0050
6.	CB-7	21.	BC-0026	36.	BC-0052
7.	CB-8	22.	BC-0027	37.	BC-0053
8.	CB-10	23.	BC-0028	38.	BC-0054
9.	CB-14 (Check)	24.	BC-0029	39.	BC-0055
10.	BC-0491	25.	BC-0033	40.	BC-0057
11.	BC-0510	26.	BC-0036	41.	BC-0058
12.	BC-0512	27.	BC-0037	42.	BC-0059
13.	JA-11/L	28.	BC-0038	43.	BC-0060
14.	JA-17/2	29.	BC-0042	44.	BC-0061
15.	JA-13/X	30.	BC-0045	45.	BC-0062

Table 4: List of the well performing 12 lines used in third experiment in 2023

Serial	Genotype
1.	RA-2
2.	RA-5
3.	RA-9.N5
4.	BC-0491
5.	BC-0510
6.	BC-0512
7.	JA-17/2
8.	JA-13/X
9.	TC-1903
10.	BC-0027
11.	CB-14 (Check)
12.	Mutant-1 (check)

3.2.4.3.2 Fertilizer Application

Fertilizers were applied at the rate of 250-175-175-100-10-10-10-6000 kg ha⁻¹ as Urea–TSP–MOP–Gypsum–Borax–Zinc sulphate–MgSO₄–Cow dung, respectively (**Table 5**), as recommended by CDB (2020–21). The total cow dung was applied at the beginning of land preparation. Basal dose (final land preparation): Urea, TSP, and MOP were applied in the rows at rates of 50, 125, and 45 kg ha⁻¹, respectively. 3 weeks after sowing (1st side dressing): Urea (50), Gypsum (30), and Zinc sulphate (10) kg ha⁻¹ were applied. 6 weeks after sowing (2nd side dressing): Urea (50), TSP (50), MOP (50), Gypsum (30), and Borax (10) kg ha⁻¹ were applied. 9 weeks after sowing (3rd side dressing): Urea (50), MOP (60), Gypsum (40), and Magnesium sulphate (10) kg ha⁻¹ were applied. 12 weeks after sowing (4th side dressing): Urea (50) and MOP (20) kg ha⁻¹ were applied.

Table 5: Rate and method of application of fertilizers and manures in the experiments

Fertilizers & Manures	Rate (kg ha ⁻¹)	Basal	3 WAS	6 WAS	9 WAS	12 WAS
Cow dung	6000	6000	–	–	–	–
Urea	250	50	50	50	50	50
TSP	175	125	–	50	–	–
MOP	175	45	–	50	60	20
Gypsum	100	–	30	30	40	–
Zinc sulphate	10	–	10	–	–	–
MgSO ₄	10	–	–	–	10	–
Borax	10	–	–	10	–	–

Source: CDB, 2020–2021

WAS = Weeks After Sowing

3.2.4.3.3 Irrigation

Experimental plots were irrigated twice. The 1st irrigation was applied in the 1st week of November and the 2nd irrigation was applied in the 3rd week of November in each season.

3.2.4.3.4 Mulching

Mulching was implemented 40 days after sowing with the objective of enhancing soil aeration and conserving moisture. These practices are known to contribute significantly

to improved plant growth and development. In instances where soil crust formation was observed, mulching was employed as a corrective measure to disrupt the crust and thereby maintain favorable soil conditions.

3.2.4.3.5 Plant Protection

Plant protection measures were initiated 30–35 days after sowing with the application of chlorpyrifos to suppress sucking insect pests, particularly jassids (*Amrasca devastans*) and aphids (*Aphis gossypii*). Subsequently, seven additional applications of chlorpyrifos in combination with pyrethroids were carried out to control both sucking and chewing insect pests, including bollworms (*Helicoverpa armigera*), spotted bollworms (*Earias* spp.), and *Spodoptera litura*. All pesticide applications were undertaken based on field scouting observations to ensure need-based intervention. Infestations of bollworms and *Spodoptera* were further managed through integrated pest management (IPM) strategies, such as mechanical removal (hand-picking) of larvae and deployment of pheromone traps to capture adult moths. These measures effectively disrupted the reproductive cycle of the pests, thereby reducing their population pressure and contributing to the maintenance of an ecologically balanced agro-ecosystem. Fungal pathogens were managed through the application of Dithane- M-45 (mancozeb) and Bavistin (carbendazim), which were sprayed thrice during the crop cycle: once at the seedling stage and subsequently after boll formation. Continuous monitoring of insect pest incidence and disease prevalence was conducted on a line-by-line basis to ensure timely and effective interventions.

3.2.4.4 Harvesting

Boll maturity was assessed when approximately 80% of the bolls had reached physiological maturity. The first harvest of seed cotton was conducted between 10–15 November, yielding approximately 50–60% of the total seed cotton. The second harvest was carried out between 25–30 November, contributing an additional 25–30% of the total yield. The remaining seed cotton was collected between 10–15 December within the cropping season. Harvested seed cottons were separately bundled, properly labeled, and transported to the threshing floor. Seed cotton yield was recorded after thorough sun-drying to ensure uniform moisture reduction.

3.2.4.5 Processing

Post-harvest processing involved careful threshing, cleaning, and sun-drying of seed cotton to estimate yield. The moisture content of seeds was adjusted to 12% to facilitate storage and preservation. The dried seed cottons were then conveyed to a ginning machine for separation of seeds and fibers.

3.2.5 Data collection

In each experiment observations were recorded from ten randomly selected plants from each genotype in each replication for the following characteristics according to guideline given by Cotton Development Board (CDB), Dhaka (**Figure 2b-e, 3a-f**).

3.2.5.1 Morphological traits

a) Plant Height (PH, cm): Plant height was measured from the base of the stem to the apex using a tape ruler in centimeter (cm).

b) Sympodial Branches per Plant (SBPP): The number of direct fruiting branches was determined by counting nodes from the base to the stem of each plant.

c) Days to 50% Flowering (DFF): It was estimated by counting the days from planting to 50% of the plants in a plot exhibited at least one open flower.

d) Days to First Boll Splitting (DFBS): It was estimated by counting the days from planting to first boll splitting.

e) Bolls per Plant (BPP): It was measured by counting total number of bolls enumerated for each plant.

f) Boll Bursting Percentage (BBP, %): It was estimated by percentage of number of burst bolls to total number of bolls and expressed in percentage (%).

g) Single Boll Weight (SBW, g): It was measured by averaging the weight of 30 selected bolls per plant in gram (g).

h) Seeds per Boll (SPB): It was measured by the number of seeds per boll, averaged across sampled bolls.

i) Seed Cotton Yield per Plant (SCYPP, g): It was measured by calculating the average yield from ten representative plants of each genotype in gram (g).



Figure 2. Showing a) field layout, b) vegetative stage, c) flowering stage, d) boll formation stage and e) boll bursting stage of upland cotton.

j) Lint Fiber Yield per Plant (LFYPP, g): It was derived by deducting the average seed weight from the total seed cotton yield, averaged across ten plants in gram (g).

k) Earliness Index (EI, %): It was calculated as the percentage of the first picking relative to total seed cotton harvest, indicating crop maturity and earliness and expressed in percentage (%).

l) Phenotypic Acceptability (PA): It was scored by visual scoring of plant vigor on a scale of 1 to 10, assessing overall vigor, phenotypic performance and robustness (higher scores indicate better performance). It was measured using scoring scale unacceptable (1), poor (3), fair (5), good (7) and excellent (9). It is a standardized qualitative measure, most notably defined within the IRRI's evaluation framework (IRRI, 2014).

3.2.5.2 Quality traits

Cotton fiber quality was comprehensively evaluated using the Engineered Fiber Selection (EFS™) system in conjunction with the High Volume Instrument (HVI®) protocols and the U.S. Cotton Fiber Chart (2023/2024).

a) Ginning Outturn (GOT, %): GOT was determined as the percentage of lint obtained from seed cotton after ginning, representing an important economic trait.

$$\text{GOT} = (\text{wt. of lint} \div \text{wt. of seed cotton}) \times 100$$

b) Upper Half Mean Length (UHML, mm): UHML was measured as the average length of the longer 50% of fibers in a sample. Fibers were categorized as short (≤ 22.0 mm), medium (22.1–24.9 mm), long (25.0–26.9 mm), and extra-long (≥ 27.0 mm).

c) Mean Length (ML, mm): ML was calculated as the weighted average of fiber lengths by summing the product of each fiber's length and weight, divided by the total fiber weight and expressed as millimeter.

d) Uniformity Index (UI, %): UI, the ratio of mean length to UHML, was expressed as a percentage. Classification thresholds included very low ($< 77\%$), low (77–79%), average (80–82%), high (83–84%), and very high ($\geq 85\%$).

e) Short Fiber Index (SFI, %): SFI indicated the proportion of fibers shorter than 12.7 mm (0.5 inches), determined using HVI® systems such as USTER.

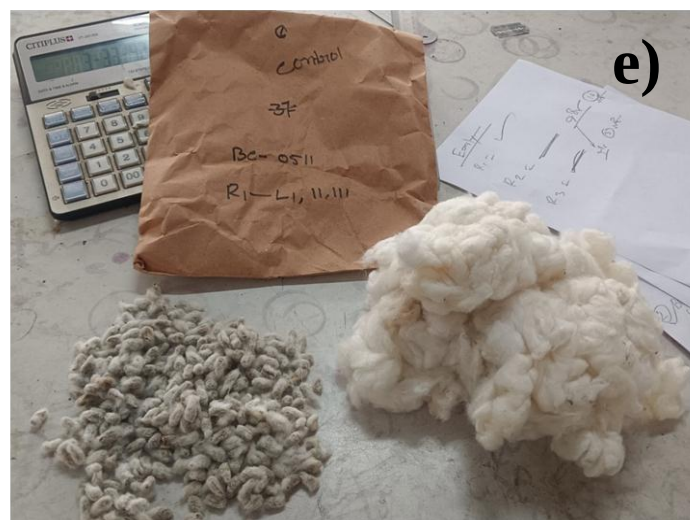


Figure 3. Measuring a) field data, b) plant height, c) seed weight, d) counting seeds per boll, e) ginning and f) lint weight of upland cotton.

f) Strength (Str., g/tex): Strength was measured as the force in grams required to break bundle of fibers one tex unit in size. Categories included weak (≤ 23 g/tex), intermediate (24-25), strong (26-29), and very strong (≥ 31).

g) Elongation (Elong., %): Elongation was recorded as the percentage increase in fiber length under tensile stress, relative to its original length. Ranges included very low ($< 5.0\%$), low (5.0–5.8%), average (5.9–6.7%), high (6.8–7.6%), and very high ($> 7.6\%$) (U.S. Cotton Fiber Chart, 2023/2024).

h) Micronaire (Mic., $\mu\text{g}/\text{inch}$): Micronaire values were used as a composite index of fiber fineness and maturity. Classes included very fine (< 135), fine (135–175), average (175–200), coarse (200–230), and very coarse (> 230) (U.S. Cotton Fiber Chart, 2023/2024).

i) Maturity Ratio (MR): Fiber maturity was expressed as a ratio using the formula:

$$\text{MR} = (\text{N}\% - \text{D}\% \div 200) + 0.7$$

Where, N represented the proportion of normal fibers and D the proportion of dead fibers. Fibers were classified as uncommon (< 0.7), immature (0.7–0.8), mature (0.8–1.0), or very mature (> 1.0) (U.S. Cotton Fiber Chart, 2023/2024).

j) Moisture (Moist., %): Lint moisture content was determined as the percentage of water present in the fiber.

k) Color Measurement (Rd and +b): Color was assessed using the HVI® system, which measures reflectance (Rd), an indicator of brightness (40 = dark, 90 = light), and yellowness (+b), an indicator of pigmentation (scale: 4-18) (HVI, U.S. Cotton Fiber Chart, 2023/2024).

l) Color Grades: Cotton samples were classified into five major groups: White, light spotted, spotted, tinged and yellow stained. Each group was subdivided into official U.S. grades, such as good middling (GM, grade 11), strict middling (SM, grade 21), middling (M, grade 31), strict low middling (SLM, grade 41), low middling (LM, grade 51), strict good ordinary (SGO, grade 61), and good ordinary (GO, grade 71). Equivalent nomenclature was applied across other groups (e.g., GM Lt Sp, Mid Sp, SLM Tg, Mid YS) (HVI, U.S. Cotton Fiber Chart, 2023/2024).

m) Cotton Grade (CG): Final cotton grade was determined by locating the intersection of Rd and +b values on the HVI® Color Chart and combining it with leaf grade, which ranges from 1 (minimal trash content) to 7 (high trash content) (HVI, U.S. Cotton Fiber Chart, 2023/2024).

3.2.6 Statistical analyses

The data obtained for different characters were recorded first on MS excel sheet. Afterwards, the data were analyzed using the software R studio of version 4.5.2 (R Core Team, 2025).

3.2.6.1 Analysis of variance (ANOVA)

Analysis of variance of morphological and fiber quality traits was performed using the package “variability” in R software version 4.5.2 (R Core Team, 2025) with the following methods:

$$y_{ij} = g_i + r_j + \epsilon_{ij}$$

Where,

Y_{ij} = Observation of genotype i in replication j

g_i = Effects of genotype i

r_j = Effects of replication j

ϵ_{ij} = The residual error of genotype i in replication j

The mean data of the genotypes were calculated by using Statistix 10 software (Statistix 10, 2025). The significance of differences among the means of genotypes were evaluated by LSD test using the package “agricolae” (De Mendiburu & Simon, 2015) in R for interpretation of results.

3.2.6.2 Estimation of post-hoc Dunnett's test

Dunnett's test uses a modified t-statistic to compare multiple treatment groups against a single control group. It was performed with the package “DescTools” in R (R Core Team, 2025). The critical value was calculated as:

$$D_{\text{Dunnett}} = t_{\text{Dunnett}} \times \sqrt{\{MSE (1 \div nc + 1 \div ni)\}}$$

Where,

t_{Dunnett} = Critical value from Dunnett's distribution,

MSE = Mean square error, and

nc = Control group sample sizes

ni = Treatment group sample sizes.

The test adjusts for multiple comparisons while maintaining overall Type I error rate.

3.2.6.3 Estimation of genotypic and phenotypic variances (σ^2_g and σ^2_p)

Genotypic and phenotypic variances were estimated according to the formula given by Johnson *et al.* (1955).

$$\text{Genotypic variance } (\sigma^2_g) = \frac{MSG - MSe}{r}$$

$$\text{Phenotypic variance } (\sigma^2_p) = \sigma^2_g + \sigma^2_e$$

Where,

MS_g = Mean sum of squares for genotypes;

MS_e = Mean sum of squares for error, and

r = Number of replications

σ^2_g = Genotypic variance, and

σ^2_e = Error mean square

3.2.6.4 Estimation of genotypic and phenotypic co-efficient of variations (GCV and PCV)

Genotypic and phenotypic co-efficient of variations were calculated according to the formula given by Singh & Chaudhary (1985); Burton (1952).

$$\text{Genotypic co – efficient of variation (GCV)} = \frac{\sqrt{\sigma^2_g}}{\bar{x}} \times 100$$

Where,

$\sqrt{\sigma^2_g}$ = Genotypic variance; and

$\bar{x}$ = Population mean

$$\text{Phenotypic co – efficient of variation (PCV)} = \frac{\sqrt{\sigma^2_p}}{\bar{x}} \times 100$$

Where,

$\sqrt{\sigma^2_p}$ = Phenotypic variance; and

$\bar{x}$ = Population mean

Here, GCV and PCV estimates were grouped as low (0–10%), moderate (10–20%), or high (20% and above) as instructed by Burton & De Vane, 1953.

3.2.6.5 Estimation of heritability (h^2_b)

The estimation of heritability in broad sense (h^2_b) was conducted following the formula proposed by Falconer & Mackay, 1996; Allard, 1960; Johnson *et al.*, 1955.

$$h^2_b(\%) = \frac{\sigma^2_g}{\sigma^2_p} \times 100$$

Where,

σ^2_g = Genotypic variance; and

σ^2_p = Phenotypic variance.

Based on description by Falconer & Mackay, 1996; Robinson *et al.*, 1949 heritability estimates were categorized as low (<30%), moderate (30-60%) and high (>60%).

3.2.6.6 Estimation of genetic advance (GA)

The estimation of genetic advance (GA) was performed utilizing the equation suggested by Allard, 1960; Johnson *et al.*, 1955.

$$\text{Genetic advance (GA)} = h^2_b \cdot K \cdot \sigma_p$$

Where,

h^2_b = Heritability in broad sense;

K = Selection intensity which is equal to 2.06 at 5% level; and

σ_p = Phenotypic standard deviation

3.2.6.7 Estimation of genetic advance as percentage of mean (GAM)

The calculation of genetic advance as a percentage of the mean (GAM) was performed using the tools developed by Comstock & Robinson, 1952; Robinson *et al.*, 1949.

$$\text{GA}(\%) = \frac{\text{GA}}{\bar{x}} \times 100$$

Where,

GA = Genetic advance; and

$\bar{x}$ = Population mean

According to Falconer & Mackay, 1996; Johnson *et al.*, 1955 GAM was grouped as low (0–10%), moderate (10–20%), or high ($\geq 20\%$).

3.2.6.8 Estimation of genotypic and phenotypic correlation co-efficient (r_g and r_p)

The correlation coefficients between the genotype and phenotype of yield and its contributing factors were evaluated by the following technique developed by Johnson *et al.*, 1955.

$$\text{Genotypic correlation of coefficient, } r_{g1.2} = \frac{\text{Cov.g1.2}}{\sqrt{\sigma^2_{g1} \times \sigma^2_{g2}}}$$

Where,

Cov.g1.2 = Genotypic covariance between the variables x_1 and x_2 .

σ^2_{g1} = Genotypic variance of the variable X_1

σ^2_{g2} = Genotypic variance of the variable X_2

Similarly,

$$\text{Phenotypic correlation of coefficient, } r_{p1.2} = \frac{\text{Cov.ph1.2}}{\sqrt{\sigma^2_{ph1} \times \sigma^2_{ph2}}}$$

Where,

Cov.ph1.2 = Phenotypic covariance between the variable X_1 and X_2

σ^2_{ph1} = Phenotypic variance of the variable X_1

σ^2_{ph2} = Phenotypic variance of the variable X_2

Here, genotypic and phenotypic correlation co-efficient were performed using the package “variability” in R software version 4.5.2 (R Core Team, 2025).

3.2.6.9 Path coefficient analysis (R)

The path coefficient analysis was determined by the technique suggested by Singh & Chaudhary, 1985; Dabholkar, 1992. This approach allowed for the separation of direct and indirect impacts on yield. Assuming 10 independent ($X_1, X_2, \dots$, and X_{10}) and one dependent variable (Y), path coefficient can be represented as follows:

$$r_{1y} = P_{1.y} + r_{1.2} P_{2.y} + r_{1.3} P_{3.y} + r_{1.4} P_{4.y} + r_{1.5} P_{5.y} + r_{1.6} P_{6.y} \dots \dots \dots r_{1.10} p_{10.y}$$

$$r_{2y} = r_{2.1} P_{1.y} + P_{2.y} + r_{2.3} P_{3.y} + r_{2.4} P_{4.y} + r_{2.5} P_{5.y} + r_{2.6} P_{6.y} \dots \dots \dots r_{2.10} p_{10.y}$$

$$r_{3y} = r_{3.1} P_{1.y} + r_{3.2} P_{2.y} + P_{3.y} + r_{3.4} P_{4.y} + r_{3.5} P_{5.y} + r_{3.6} P_{6.y} \dots \dots \dots r_{3.10} p_{10.y}$$

$$r_{4y} = r_{4.1} P_{1.y} + r_{4.2} P_{2.y} + r_{4.3} P_{3.y} + P_{4.y} + r_{4.5} P_{5.y} + r_{4.6} P_{6.y} \dots \dots \dots r_{4.10} p_{10.y}$$

$$r_{5y} = r_{5.1} P_{1.y} + r_{5.2} P_{2.y} + r_{5.3} P_{3.y} + r_{5.4} P_{4.y} + P_{5.y} + r_{5.6} P_{6.y} \dots \dots \dots r_{5.10} p_{10.y}$$

.....

$$r_{16y} = r_{16.1} P_{1y} + r_{16.2} P_{2y} + r_{16.3} P_{3y} + r_{16.4} P_{4y} + r_{16.5} P_{5y} \dots r_{10.16} P_{10y}$$

Where,

$P_{1y}, P_{2y}, P_{3y}, \dots, P_{10y}$ = Path coefficient of the variables $x_1, x_2, x_3, \dots, x_{10}$ on the variable Y respectively.

After calculating the direct and indirect effects of the character's residual effects (R) was calculated as follows:

$$\text{Residual effect, } R = \sqrt{1 - (r_{1y}P_{1y} + r_{2y}P_{2y} + \dots + r_{10y}P_{10y})}$$

Here, the values of direct and indirect effects of path-coefficient were scaled as negligible (0.00 to 0.09), low (0.10 to 0.09), moderate (0.20 to 0.29), high (0.30 to 0.99) and very high (<1.00). The path co-efficient was performed using the package “variability” in R software version 4.5.2 (R Core Team, 2025).

3.2.6.10 Diversity analysis through multivariate analytical methods

Multivariate analytical methods, such as cluster analysis and principal component analysis (PCA), are effective instruments for evaluating genetic diversity by quantifying differences among different quantitative variables (Brown-Gudera *et al.*, 2000) which were used in the current study.

3.2.6.10.1 Cluster analysis

Pooled mean data were employed for clustering with Python Program (Pilgrim & Willison, 2009). A hierarchical agglomerative approach was employed, utilizing Euclidean distances in Ward's D method (Ward, 1963). The clustering technique was designed to minimize within-group variance, hence earning the designation Ward's minimum variance method. The supreme number of clusters was identified at the juncture where the overall within-cluster variance demonstrated a substantial decrease. Python programming language V 3.9.7 (Python, 2023) was used for estimating cluster analysis.

3.2.6.10.2 Principal component analysis (PCA)

The technique was used to examine the interrelationships among the traits. The principal components were computed from the correlation matrix obtained from the sum of squares product matrix of the characters and the genotype score obtained from the first component and the succeeding component with latent roots greater than unity. The latent roots are called Eigen values. The component has the property of accounting for

maximum variances. The PCA displays most of the original variability in a smaller number of dimensions since it finds linear combinations of a set of varieties that maximize the variation contained within them. Contributions of the different characters towards divergence are discussed from the latent vectors of the first two principal components. The coefficients of the proper vector exceeding 0.3, irrespective of positive or negative direction, indicate significant effects deemed important, whereas traits below 0.3 lack substantial impact on overall variation (Raji, 2003). PCA was performed in R statistics version 4.5.2 (R Core Team, 2025).

3.2.6.10.3 Biplot analysis

A PCA biplot is a two-dimensional chart that shows both the PC scores of samples and the loading of variables. By using the relative value of the trait, a genotype $\times$ trait biplot was constructed from a two-way matrix of traits and genotypes using “Factoextra” and “FactoMineR” packages in R software version 4.5.2 (Kassambara & Mundt, 2017).

3.3 Molecular characterization of upland cotton genotypes

3.3.1 Plant materials

Ten outstanding short durated genotypes viz., RA-2, RA-5, RA-9.N5, BC-0491, BC-0510, BC-0512, JA-17/2, JA-13/X, TC-1903, BC-0027 with two popular checks viz., CB-14 and Mutant-1 selected from 2nd year were used as plant genetic materials for molecular characterization (**Figure 4**).

3.3.2 Experimental location and tenure

The molecular work was conducted in July-December, 2023 in the Genetics and Plant Breeding Lab, and Central Lab. of Hajee Mohammad Danesh Science and Technology University, Dinajpur, Bangladesh (**Figure 4**).

3.3.3 Genomic DNA isolation

The DNA was extracted from 14 days old seedlings. Here, about 2-4 pieces of young leaves were collected in Eppendorf tube and were dried for 2 weeks in silica gel (**Figure 5**). The samples were ground using mortar and pestle. Afterwards, 800 μ l of warmed (65°C) CTAB buffer was added to each tube and was vortexed thoroughly. Samples were incubated in water bath at 65°C for 45 minutes and at every 10 minutes they were mixed gently by inversion (400 μ l of 2% β -Mercaptoethanol was added to 200 ml of extraction



Figure 4. Molecular study of upland cotton showing a) seeds of the plant materials, b) growing into petri dishes, c) young seedlings of the selected materials, d) preparing samples for PCR and e) loading samples into PAGE.

buffer prior to warming). The tubes were taken out of the water bath and left at room temperature for 5 minutes. 600 µl chloroform Isoamyl alcohol was added (24:1). The samples were mixed by gently inversion for about 2 minutes (100 times) until two layers' mix. Then the samples were centrifuged for 4000 rpm at room temperature for 20 mins. The aqua phase was removed with wide bore pipette. The aqua phase was transferred to clean 1.5 ml tube then 2/3 volume of Ethanol was added and mixed gently to precipitate the nucleic acids. At this stage the samples were stored in 4°C for overnight. After that the samples were centrifuged at 10000 rpm for 20 minutes and the supernatant was discarded. Then the DNA was dried so that there was no ethanol. A volume, 500 µl of washing buffer was added in each tube and the DNA was washed gently inversion. Again, the samples were centrifuged at 10000 rpm for 20 mins, then the supernatant was removed and the tubes were left on bench for drying. 100 µl double distilled water was added, the samples were left for dissolving. Finally, the samples were stored in 4°C for short term (**Figure 5**).

3.3.4 DNA quantification

The Quality of the extracted DNA samples were also checked prior to PCR amplification through quantification using a Thermo Scientific Nano Drop[^] TM1000 spectrophotometer (Thermo Fisher Scientific, USA) (**Figure 6**).

3.3.5 PCR amplification and electrophoresis separation

PCR amplifications were performed in 10 µL tube using a Veriti Thermal Cycler (Applied biosystems, USA). 2µL of template DNA, and 8 µL (0.5 µL of Forward primer, 0.5µL of Reverse primer, 2µL of nuclease free water and 5µL of G2 Green Master Mix) of reaction mixture was added in each tube. The PCR amplification was as follows: one cycle of 94°C for 5 min; 35 cycles of 95°C for 0.5 min, 53 to 58°C (depending on the specific primers) for 0.5 min and extension for 0.5 min; and a final extension at 72°C for 5 min. Reaction products were mixed with one fifth volume of loading buffer (100 mM/L EDTA pH 8.0, 10 mM/L Tris-HCl pH 7.5, 5% Ficoll 400; 0.05% bromophenol, 0.05% xylene cyanol) and 2 µL were loaded vertically, for electrophoresis 8% denaturing polyacrylamide gels in 1 × TBE (90mM/L Tris borate pH 8.3, 2 mM/L EDTA) at 50 mA for 2 to 3 h (Wang *et al.* 2007). Gels were then silver stained and photographed using x-ray viewer (**Figure 6**).

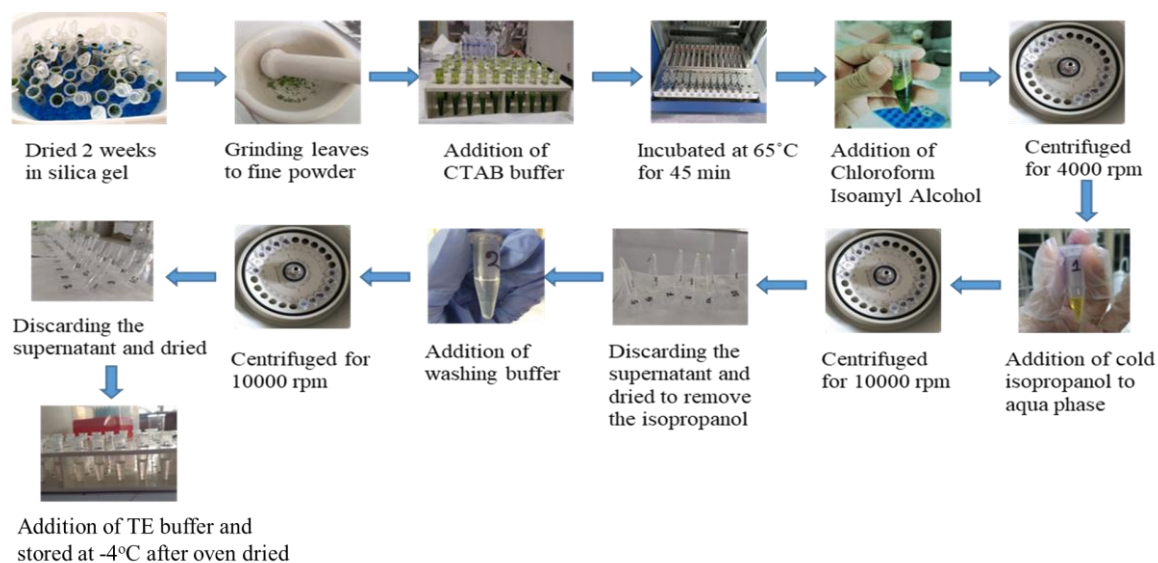


Figure 5. DNA extraction using modified CTAB method

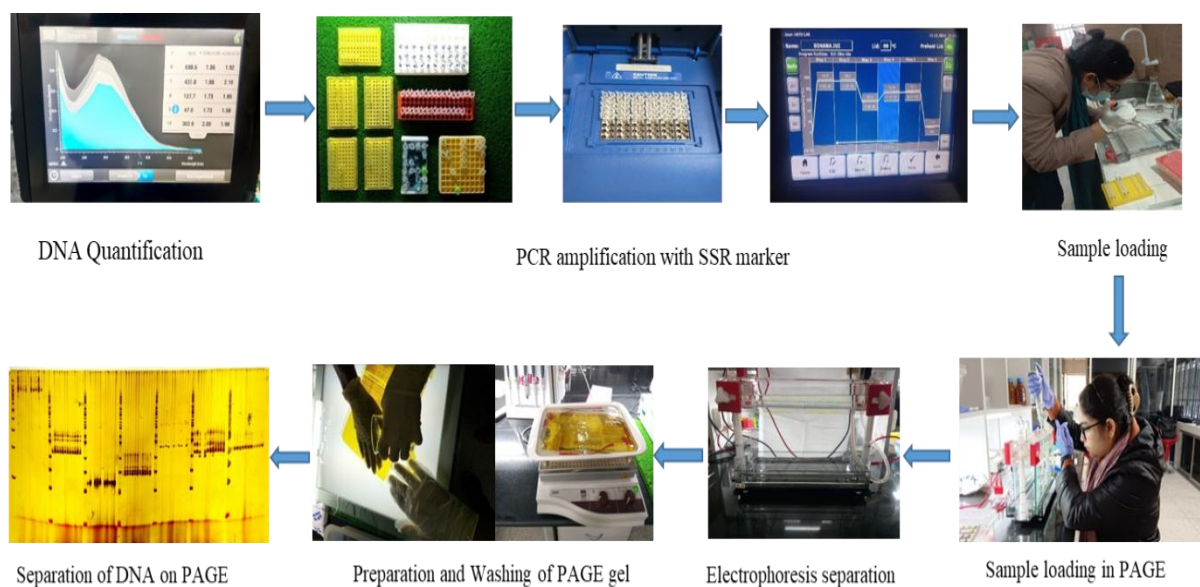


Figure 6. PCR amplification and electrophoresis separation

3.3.4 Microsatellite/Simple sequence repeat (SSR) markers

A total of 20 microsatellite (SSR) markers primer pairs (sigma Aldrich, Germany) were used to characterize 12 upland cotton genotypes as shown in the **Table 6**. These SSR primers with a distinct chromosome numbers were used for final Polymerase Chain Reaction (PCR) amplification. The original sources, repeat motifs, primer sequences, expected length and chromosomal position and other relevant information were retrieved from Çelik, 2022. The chromosomal location, annealing temperature and primer sequences to SSR markers are shown in **Table 6**.

3.3.5 Molecular Statistical Analysis

Polymorphism information content (PIC) were calculated using the following formula:

$$PIC= 1-\sum (P_i)^2$$

Here, P_i depicted the proportion of samples carrying the i th allele.

Model-based clustering program STRUCTURE V 2.3.4 (Pritchard *et al.* 2000) was employed to deduce the population structure of all the 12 upland cotton genotypes. Number of populations (K) was determined with a burn-in period of 100000 and Markov Chain Monte Carlo of 100000. Five independent runs were performed for each K varying from 1 to 10. In this model, several subpopulations (K) were assumed to be present, each of which was resembled a set of allele frequencies for each locus. Individuals in the sample were assigned to subpopulations or jointly to two or more subpopulations if their genotypes were admixed. The model choice criterion to detect the most likely value of K was ΔK , an ad hoc quantity related to the second-order change in the log probability of data ($\ln P[D]$) with respect to the number of clusters inferred by Structure (Evanno *et al.* 2005). An individual was assigned to a subpopulation group if >80% of its genome fraction value was derived from that group. Most probable K-value was defined based on DK method (Evanno *et al.* 2005) by running the Structure Harvester software (Earl & VonHoldt, 2012). Every band was considered as a single locus. All the scorable loci were considered for generation of bivariate 1-0 data matrix and were used for estimation of dendrogram by Metabo Analyst (Online Version) software (Chong & Xia, 2018).

Table 6. SSR markers with primer sequences and annealing temperature used in well performing 12 upland cotton genotypes

SL.	SSR Loci	Forward primer (5'–3')	Reverse primer (5'–3')	Annealing temperature (°C)
1.	BNL1604	AGAGGGAGTAAAGATTTGGGG	TCCAGTTCTTTTGCCTTGG	54.44
2.	BNL1611	CAATGAACAAAAAATGTAAGGG	TGGGCATTTAGCCATTACC	51.85
3.	BNL3449	AAGCTGTGGCTATGATGCCT	AGAGCAAAAAACAATTACAAAAGC	53.32
4.	BNL3479	AGTGGGTTGGACTTTCATGC	CACGGGCTTTTTTTTTTTTCA	52.23
5.	NAU3736	CATGTGCATTTTCATCCTGTC	CCAAGTGAGAGGCATTTTCT	53.25
6.	NAU4024	ACAAGCATCTTCATGGACCT	AGAAGGATGATGCAAAGAGG	53.25
7.	MGHES16	ACCCCAATACAACCCCATTT	GCAGAGAAAAGGGACAGAGG	55.3
8.	FPF1	AAACTCAGGTTTCGGACCAAAGG	ACCAAGAGACAACAACCTTAGTC	55.54
9.	Gorai.ATC	ATGGCAAAACTGTCAGATCCTCT	TTAGCGTCTTCTAGCAGCTGTTTC	57.20
10.	BNL2571	TCGCTATCGCTCTGAAATCA	ATGCCACGGAATTAGCAAAC	53.25
11.	NAU2173	GCCAAATAGGTCACACACAA	AGCGAGAAGGAGACAGAAAA	53.25
12.	NAU2251	TTCTCCAGTAACCAACAAAGG	AAAATATCATCCCCGTCAA	51.42
13.	NAU2714	GCAGCCATTACAGAACATCA	TCATTGATCCATTGCTTCTG	52.23
14.	NAU2761	GACAAGTTTTTGGACCCACT	TTCATAGAGGGGTTTTGCAT	52.23
15.	CIR0141	CGCACAAGGAATAGAAG	ACCCAACATAAGGACTAAA	51.00
16.	CIR0253	CCAACCAAGAAACCAG	GTAAGCATGGGCATTT	47.00
17.	JESPR0065	CCACCCAATTTAAGAAGAAATTG	GGTTAGTTGTATTAGGGTCGTTG	54.43
18.	JESPR0114	GATTTAAGGTCTTTGATCCG	CAAGGGTTAGTAGGTGTGTATAC	53.71
19.	JESPR0153	GATTACCTTCATAGGCCACTG	GAAAACATGAGCATCCTGTG	54.44
20.	Gohir	ATGTCGTGCATGTCAAATGG	TACAGCACGTACAGTTTACC	53.25

Source: The original sources, repeat motifs, primer sequences, expected length and chromosomal position and other relevant informations were retrieved from Çelik, 2022.

CHAPTER IV

RESULTS AND DISCUSSION

Cotton is one of the most economically important commercial crops, serving as the principal natural fiber for the global textile industry and widely regarded as the “King of Fibre Crops.” In Bangladesh, all available and cultivated varieties are in general low fiber yield potential, not exceeding 3500kg/ha but world’s bench mark fiber yield is 4000kg/ha. Cotton productivity remains low, largely due to prolonged crop duration, low yield potentials, the predominance of rainfed cultivation (>75%), limited use of quality seed, sub-optimal nutrient management, and inadequate plant protection practices. These constraints collectively limit yield potential and highlight the need for improved genetic and agronomic strategies. Accordingly, the present research aimed to identify early boll-picking, high-yielding upland cotton genotypes to enhance national cotton productivity.

4.1 Experiment I: Characterization of 100 upland cotton (*Gossypium hirsutum* L.) genotypes to select fiber yield enhancing traits and promising early genotypes

The major goal of cotton breeders is to develop high yielding modern cotton varieties with promising characteristics in terms of early maturing, short boll picking period, phenotypic acceptability etc. It will definitely increase farmer’s profitability by cutting extra budget for conventional long-term cotton cultivation in field. The attainment of this objective is obstructed by the diminished and narrowed genetic base of contemporary cotton varieties, resulting from the ongoing massive selection process, which has ultimately led to a deficiency in genetic variability among the core cotton genotypes. Thus, the present study aimed to assess the genetic variability and diversity across the 100 upland cotton genotypes including elite lines, popular cultivars, mutant derivatives, and advanced breeding lines selected for their varied morphological and yield-related traits.

4.1.1 Analysis of variance (ANOVA) and descriptive statistics of 100 upland cotton genotypes

The analysis of variance and descriptive statistics for eleven morphological traits of 100 upland cotton genotypes are presented in **Table 7**. The variance analysis revealed significant differences among the genotypes across all traits indicating substantial genetic variability, which included phenotypic acceptability, plant height (cm), sympodial branches per plant,

days to 50% flowering, days to first boll splitting, bolls per plant, single boll weight (g), seeds per boll, seed cotton yield per plant (g), lint fiber yield per plant (g), and earliness index (%). Analogous results were obtained previously (Sarwar *et al.*, 2021a; Nivedha *et al.*, 2020). Consequently, there exists a significant possibility to choose superior parental genotypes for breeding exercise to enhance seed cotton output. The genotype mean sum of squares values was significantly larger than the error mean sum of squares for all characteristics revealed good precision in analysis of variance for eleven traits.

The replication effects were largely non significant for most traits, except for plant height and days to 50% flowering due to minor heterogeneity of in the blocks. The coefficient of variation varied from 1.05% for seed cotton yield per plant to 8.64% for phenotypic acceptability. There were wide ranges of variations revealed for the mean performances of all studied traits. The averages of earliness traits, specifically days to 50% flowering and days to first boll splitting, were 48.54 ± 0.22 days and 130.44 ± 0.30 days, respectively. The average days to 50% flowering was reported in other findings as 72.48 days (Raj & Patil, 2023), 60.67 days (Saraswat *et al.*, 2022) and 65.04 days (Sarwar *et al.*, 2021b). Our findings reported at at least 12 days earlier flowering than others that will be helpful to develop early variety in Bangladesh. The current study acknowledged that phenotypic acceptability ranged from 2.80 to 9.60, with the largest coefficient of variation (CV) at 8.64%. Raj & Patil, (2023) reported 15.32% earliness index whereas 57.73% earliness index has also been stated (Mahdi & Emam, 2020) in cotton. This study revealed higher mean (>80%) of earliness index that will increase the chance of the genotypes to include earliness in cotton variety development.

There were a wide range of variations for plant height (cm), number of sympodial branches per plant, bolls per plant, single boll weight (g), seeds per boll in 100 upland cotton genotypes of the current study. Salahuddin *et al.*, (2010) argued that this demonstrated substantial genetic variability of the attributes among the genotypes, which has further impacted other associated traits. Amer & Hassan, 2023; Amer, 2022; Sahar *et al.*, 2021; Sarwar *et al.*, 2021a; Chapepa *et al.*, 2020; Rehman *et al.*, 2020; Kumar *et al.*, 2019 also found significant differences among the cotton genotypes. The wide range among cotton genotypes for the different plant characters were also outlined by Yasar, 2023; Balarabe *et al.*, 2022; Amer *et al.*, 2021; Gibely, 2021; Bhatti *et al.*, 2020a,b; Nivedha *et al.*, 2020; Sarwar *et al.*, 2020; Jarwar *et al.*, 2018.

Table 7. Mean squares (MS) derived on 11 morphological characters in 100 upland cotton genotypes

Character	Mean Sum of Squares					
	Genotype	Replication	Error	CV (%)	Mean Range	Grand Mean $\pm$ SE
Phenotypic acceptability	4.89***	0.21	0.29	8.64	2.80-9.60	6.18 $\pm$ 0.13
Plant height (cm)	1901.85***	77.11*	17.16	3.12	83.81-181.67	132.69 $\pm$ 2.52
Sympodial branches per plant	5.09***	0.08	1.22	5.77	15.60-24.20	19.17 $\pm$ 0.13
Days to 50% flowering	14.09***	3.69*	0.96	2.02	43.20-54.80	48.54 $\pm$ 0.22
Days to first boll splitting	26.07***	1.68	2.29	1.16	123.20-141.20	130.44 $\pm$ 0.30
Bolls per plant	41.04***	0.91	3.92	7.72	12.20-36.20	25.67 $\pm$ 0.37
Single boll weight (g)	0.99***	0.06	0.09	6.41	3.45-6.57	4.69 $\pm$ 0.06
Seeds per boll	14.86***	7.02	3.41	6.02	21.80-38.20	30.70 $\pm$ 0.22
Seed cotton yield per plant (g)	11.53***	4.73	1.64	1.05	112.33-129.00	122.59 $\pm$ 0.20
Lint fiber yield per plant (g)	15.94***	4.15	1.57	1.10	100.48-121.55	114.43 $\pm$ 0.23
Earliness index (%)	22.97***	12.04	5.42	2.72	75.12-94.92	85.57 $\pm$ 0.28

Here, * and *** indicated significant at 5% and 0.1% levels of probability, respectively. CV= coefficient of variation and SE= standard error.

Similarly, numerous prior investigations have exhibited analogous trends in the evaluated agro-morphological parameters (Çakmak *et al.*, 2023; Saraswat *et al.*, 2022; Sarwar *et al.*, 2021a).

The present study conceded that the mean of seed cotton yield was 122.59 g which ranged from 112.33-129 g per plant (**Table 7**). The mean of seed cotton yield per plant was estimated 48.67 g which varied between 31.17 and 53.22 g per plant (Mahdi & Emam, 2020). The mean of seed cotton yield per plant was estimated 70.08 g which ranged from 15.75-135.78 g (Rathinavel, 2018). Therefore, our study revealed high seed cotton yielding genotypes of upland cotton.

The phenotypic acceptability ranged from 2.80-9.60 with a mean of 6.18 ± 0.13 . The maximum variation revealed for the character plant height where the range was 83.81-181.67 cm with a mean of 132.69 ± 2.52 cm. The minimum variation exhibited for the character single boll weight where the range was 3.45-6.57 g with a mean of 4.69 ± 0.06 g. The important yield contributing character bolls per plant ranged from 12.20-36.20 with a mean of 25.67 ± 0.37 . The seed cotton yield ranged from 112.33-129.00 g per plant with a mean of 122.59 ± 0.20 g per plant whereas the lint fiber yield ranged from 100.48-121.55 g per plant with a mean of 114.43 ± 0.23 g per plant. The earliness index showed the range of 75.12% to 94.92% with a mean of $85.57 \pm 0.28\%$.

4.1.2 Mean performances of yield and its related characters among 100 upland cotton genotypes

The average performances of eleven yield and its attributing traits of 100 upland cotton genotypes are displayed in **Table 8**. Mean performance revealed, the genotype BC-0062, RA-5, BC-0510, BC-0036 had excellent phenotypic acceptability of 9.2 and 9.0 respectively. In contrast, CB-8, BC-0049 and BC-0042 had poor phenotypic acceptability of 3.0 and 3.3.

The mean plant height at the time of first boll picking varied between 86.87 cm and 177.47 cm. The genotype CB-10 was the tallest plant, measuring 177.47 cm followed by CB-8 (175.48 cm) and BC-0025 (175.34 cm) while the genotype BC-0397 was the shortest plant, measuring 86.87 cm followed by BC-0043 (88.29 cm) and BC-0513 (89.98 cm), respectively. Whereas, Mawblei *et al.*, (2022) found plant height ranged from 56-103 cm. Bhatti *et al.*, (2020a,b) measured plant height ranged from 98 to 150 cm.

Sympodial branches per plant ranged from 16 to 23.18. The genotype BC-0057 had the highest number of sympodial branches per plant (23.18) whereas, the genotype BC-0036 and BC-0052 had the minimum measuring the value 16.00. An increased number of sympodial branches per plant implies impact the effectiveness of flowering and boll formation, which in turn enhances growth and yield. The results were congruent with Mawblei *et al.*, (2022) found that, the genotype D16 showed the highest performance for sympodia per plant (20.3) among 100 different genotypes. Rehman *et al.*, (2020) reported that the genotype VH-367 exhibited the maximum mean value of 23.13 subsequent to A555 (22.66), IUB-222 (20.93) and NIAB-414 (20.93), respectively for sympodial branches per plant.

Days to 50% flowering is an important character to determine earliness in cotton hence an important aspect for cotton breeders to incorporate this trait to develop early boll setters in cotton. In the present study, the genotype BC-0024, BC-0043 and RA-2, BC-0027 were the early flowering genotypes comprised of 44.33 days and 44.67 days to 50% flowering respectively whereas, the late flowering genotypes were BC-0061, BC-0062 and CB-4, BC-0495 comprised of 54 days and 52.73 days to 50% flowering, respectively. Bhatti *et al.*, (2020 a,b) found that days to 50% flowering varied from 61-70 days, however the lowest and statistically at par value was expressed by the cultivar FH-142 followed by MNH-886. Therefore, the present study revealed justification of early flowering assessment in upland cotton genotypes.

Days to first boll splitting varied between 125-140 days. The early boll splitting genotypes CB-7 and BC-0397 comprised of 125 days and 126 days while the late boll splitting genotypes BC-0033 and BC-0036, BC-0053 comprised of 140 days, 138 days and 138 days, respectively (**Table 8**).

Among other yield related attributes bolls per plant is very important feature associated with high yielding cultivars and discussed by many reseachers. The maximum bolls per plant bearing genotypes were BC-0033, BC-0062 and BC-0055 comprised of 34.33, 34.0 and 33.0 bolls per plant. Mawblei *et al.*, (2022) investigated bolls per plant in 100 different genotypes, where, Stardel (23.67) showed the highest performance for bolls per plant. Bhatti *et al.*, (2020 a,b) opined that bolls per plant varied from 13 bolls to 31 on the basis of variation among genotypes. Rehman *et al.*, (2020) reported that bolls per plant varied from 16.26 to 32.13.

Table 8. Mean values based on phenotypic expressions of 11 morphological characteristics in 100 upland cotton genotypes

SL.	Genotype	PA	PH (cm)	SBPP	DFE	DFBS	BPP	SBW (g)	SPB	SCYPP (g)	LFYPP (g)	EI (%)
1	RA-2	7.00 ^{g-l}	155.47 ^{e-i}	19.00 ^{f-p}	44.67 ^c	136.00 ^{b-d}	28.00 ^{e-j}	4.37 ^{u-h}	36.00 ^{ab}	122.24 ^{h-a}	114.36 ^{n-f}	91.65 ^{ab}
2	RA-4	6.67 ^{h-o}	143.65 ^{l-p}	17.87 ^{n-s}	50.53 ^{d-k}	127.67 ^{u-a}	32.00 ^{a-d}	5.83 ^{b-d}	32.47 ^{c-l}	121.58 ^{e-d}	113.32 ^{v-i}	84.55 ^{g-s}
3	RA-5	9.00 ^{ab}	157.21 ^{d-h}	17.80 ^{n-s}	47.33 ^{r-x}	132.00 ^{g-m}	32.00 ^{a-d}	3.83 ^{i-l}	31.00 ^{f-s}	122.51 ^{k-y}	115.13 ^{g-x}	86.45 ^{c-n}
4	RA-9.N5	7.00 ^{g-l}	150.82 ^{h-k}	19.00 ^{f-p}	51.33 ^{b-f}	128.00 ^{a-a}	28.00 ^{e-j}	4.46 ^{r-f}	30.00 ^{j-t}	124.87 ^{a-f}	114.20 ^{o-f}	81.61 ^{r-u}
5	RA-15	6.67 ^{h-o}	151.56 ^{f-k}	19.20 ^{e-p}	47.33 ^{r-x}	129.33 ^{a-x}	32.53 ^{a-c}	5.64 ^{c-e}	32.67 ^{c-k}	121.50 ^d	113.23 ^{x-j}	87.28 ^{c-l}
6	RA-16	5.87 ^{o-v}	171.51 ^{ab}	20.40 ^{c-h}	44.80 ^{bc}	133.73 ^{d-h}	20.00 ^{b-e}	4.63 ^{l-b}	33.73 ^{b-f}	122.89 ^{f-x}	112.69 ^{c-j}	87.98 ^{b-i}
7	SR-17	7.00 ^{g-l}	158.12 ^{d-f}	18.53 ^{t-r}	47.00 ^{t-z}	128.00 ^{a-a}	25.87 ^{h-r}	5.46 ^{d-g}	32.40 ^{c-l}	123.44 ^{c-u}	108.71 ^l	83.45 ^{m-t}
8	SR-18	4.75 ^{yz}	148.01 ^{j-n}	17.93 ^{m-s}	48.13 ^{n-u}	130.33 ^{k-s}	26.80 ^{g-n}	4.91 ^{i-s}	31.80 ^{d-p}	122.68 ^{h-y}	113.15 ^{x-j}	88.56 ^{b-d}
9	SR-19	7.40 ^{e-h}	116.93 ^{z-d}	17.87 ^{n-s}	51.00 ^{c-g}	129.20 ^{a-x}	30.13 ^{c-f}	4.49 ^{r-e}	31.87 ^{d-o}	123.68 ^{c-q}	115.49 ^{d-u}	87.48 ^{c-k}
10	CB-1	4.53 ^z	127.72 ^{uv}	19.73 ^{d-l}	47.67 ^{p-w}	128.93 ^{y-y}	27.07 ^{fm}	4.46 ^{r-f}	31.53 ^{d-r}	123.92 ^{c-o}	115.76 ^{b-p}	85.99 ^{c-p}
11	CB-2	5.00 ^{w-z}	162.71 ^{cd}	20.00 ^{d-j}	50.33 ^{d-k}	130.67 ^{t-r}	23.00 ^{b-b}	5.38 ^{d-l}	30.00 ^{j-t}	123.65 ^{c-q}	115.34 ^{f-u}	78.23 ^{uv}
12	CB-3	5.93 ^{o-u}	167.41 ^{bc}	20.73 ^{b-f}	51.60 ^{b-e}	130.33 ^{k-s}	24.73 ^{k-w}	4.99 ^{g-p}	29.20 ^{n-u}	123.5 ^{c-t}	114.88 ^a	86.41 ^{c-n}
13	CB-4	6.20 ^{j-s}	154.68 ^{e-j}	19.27 ^{d-o}	52.73 ^{ab}	132.20 ^{g-l}	25.27 ^{j-u}	4.86 ^{j-t}	33.53 ^{b-g}	122.10 ^{b-b}	114.98 ^{i-z}	85.85 ^{c-p}
14	CB-5	6.27 ^{k-s}	173.34 ^{ab}	19.27 ^{d-o}	52.13 ^{bc}	130.47 ^{k-r}	28.80 ^{e-i}	4.43 ^{s-g}	30.27 ^{i-t}	121.64 ^{a-d}	111.89 ^{h-k}	79.72 ^{v-n}
15	CB-6	5.00 ^{w-z}	160.17 ^{de}	19.00 ^{f-p}	47.67 ^{p-w}	127.00 ^{b-b}	20.73 ^{a-d}	6.14 ^{ab}	34.00 ^{b-e}	122.81 ^{f-x}	114.67 ^{d-l}	86.45 ^{c-n}
16	CB-7	5.00 ^{w-z}	173.63 ^{ab}	20.00 ^{d-j}	47.33 ^{r-x}	125.00 ^b	23.00 ^{b-b}	5.35 ^{d-i}	34.27 ^{a-d}	123.16 ^{d-w}	115.35 ^{f-u}	84.88 ^{d-s}
17	CB-8	3.00 ^a	175.48 ^a	20.00 ^{d-j}	50.33 ^{d-k}	130.00 ^{l-u}	19.67 ^{c-e}	6.44 ^a	35.00 ^{a-c}	120.32 ^{z-d}	112.37 ^{f-k}	84.88 ^{d-s}
18	CB-9	4.80 ^{yz}	153.28 ^{f-j}	19.73 ^{d-l}	50.67 ^{c-j}	131.40 ^{b-o}	24.13 ^{m-y}	5.06 ^{f-m}	28.13 ^{s-v}	122.55 ^{j-y}	114.83 ^{i-b}	86.53 ^{c-n}
19	CB-10	7.00 ^{g-l}	177.47 ^a	20.00 ^{d-j}	49.33 ^{i-o}	127.00 ^{b-b}	28.00 ^{e-j}	4.33 ^{w-h}	32.00 ^{d-n}	121.26 ^{v-d}	114.43 ^{m-e}	85.78 ^{c-p}
20	CB-11	6.20 ^{j-s}	172.90 ^{ab}	18.47 ^{t-r}	47.27 ^{r-y}	130.93 ^{i-q}	23.00 ^{b-b}	4.76 ^{k-y}	29.00 ^{o-u}	121.89 ^{o-d}	111.24 ^k	88.28 ^{b-g}
21	CB-12	6.93 ^{g-m}	148.03 ^{j-n}	19.93 ^{d-k}	45.13 ^{a-c}	131.33 ^{p-p}	25.47 ^{j-t}	4.50 ^{q-d}	32.40 ^{c-l}	122.37 ^{l-z}	115.36 ^{f-u}	85.77 ^{c-p}
22	CB-13	6.40 ^{j-r}	157.95 ^{d-g}	20.80 ^{b-e}	45.73 ^{y-c}	129.13 ^{a-x}	28.80 ^{e-i}	4.68 ^{l-z}	32.47 ^{c-l}	121.98 ^{a-c}	112.49 ^{e-k}	83.00 ^{n-t}
23	CB-14	7.00 ^{g-l}	139.76 ^{o-r}	23.00 ^a	48.00 ^{o-v}	126.67 ^{b-b}	27.00 ^{f-m}	4.53 ^{p-c}	29.00 ^{o-u}	122.37 ^{l-z}	111.74 ^{i-k}	86.45 ^{c-n}
24	CB-15	6.13 ^{m-t}	121.02 ^{w-a}	19.47 ^{d-o}	48.73 ^{l-r}	130.93 ^{i-q}	25.27 ^{j-u}	4.08 ^{c-l}	31.13 ^{r-r}	122.29 ^{m-a}	113.95 ^{p-g}	85.60 ^{c-q}
25	CB-16	6.07 ^{n-t}	99.16 ^{l-o}	19.07 ^{e-p}	45.13 ^{a-c}	130.53 ^{k-r}	23.93 ^{m-z}	4.46 ^{r-f}	30.73 ^{g-t}	122.78 ^{g-x}	113.53 ^{s-i}	82.91 ^t
26	CB-17	5.73 ^{p-w}	135.58 ^{q-s}	17.80 ^{n-s}	45.27 ^{a-c}	129.13 ^{a-x}	23.27 ^{o-a}	4.43 ^{s-g}	31.53 ^{d-r}	123.95 ^{c-o}	114.99 ^{i-z}	84.31 ^s
27	CB-1055	5.87 ^{o-v}	102.34 ^{k-n}	19.33 ^{d-o}	45.87 ^{x-c}	127.60 ^{a-u}	22.20 ^{u-c}	4.22 ^{z-j}	32.40 ^{c-l}	121.66 ^{q-d}	113.27 ^{w-d}	87.37 ^{c-k}
28	BC-0385	5.07 ^{v-z}	90.01 ^{q-s}	17.47 ^{p-t}	49.20 ^{j-p}	129.53 ^{a-w}	22.53 ^{s-c}	4.55 ^{p-c}	32.80 ^{c-j}	121.78 ^{b-d}	113.48 ^{u-i}	87.99 ^{b-i}
29	BC-0397	6.00 ^{o-u}	86.87 ^s	18.53 ^{t-r}	45.60 ^{z-c}	125.67 ^{ab}	22.07 ^{v-c}	5.04 ^{f-o}	29.07 ^{n-u}	124.35 ^{a-m}	114.71 ^{j-c}	86.32 ^{c-o}
30	BC-0410	6.97 ^{g-n}	95.64 ^{o-q}	19.67 ^{d-m}	50.13 ^{e-l}	129.07 ^{o-y}	25.53 ^{j-s}	5.07 ^{f-m}	30.33 ^{h-t}	123.40 ^{c-u}	117.26 ^{a-f}	87.59 ^{c-k}
31	BC-0415	6.40 ^{j-r}	108.75 ^{g-k}	18.47 ^{t-r}	50.20 ^{e-l}	133.20 ^{e-i}	25.40 ^{j-t}	4.21 ^{z-j}	31.73 ^{d-p}	124.45 ^{a-k}	118.45 ^a	87.59 ^{c-k}
32	BC-0419	5.93 ^{o-u}	111.12 ^{d-j}	18.40 ^{t-r}	49.13 ^{j-p}	132.13 ^{g-l}	27.47 ^{f-l}	4.31 ^{x-i}	32.27 ^{c-m}	122.79 ^{g-x}	115.28 ^{f-w}	84.75 ^{e-s}
33	BC-0435	6.53 ^{i-p}	108.84 ^{f-k}	18.93 ^{g-p}	49.00 ^{k-q}	128.33 ^z	25.40 ^{j-t}	4.42 ^{t-g}	29.07 ^{n-u}	124.70 ^{a-h}	117.13 ^{a-g}	84.28 ^s
34	BC-0436	5.33 ^{t-z}	109.64 ^{e-j}	18.87 ^{h-q}	49.93 ^{f-l}	130.93 ^{i-q}	25.47 ^{j-t}	4.69 ^{l-z}	31.27 ^{e-r}	122.20 ^{n-a}	113.03 ^{z-j}	87.11 ^{c-m}
35	BC-0442	5.47 ^{s-y}	107.94 ^{h-k}	18.80 ^{h-q}	51.67 ^{b-e}	132.00 ^{g-m}	24.33 ^{l-x}	4.33 ^{v-h}	28.60 ^{r-v}	123.65 ^{c-q}	115.41 ^{e-u}	84.79 ^{e-s}
36	BC-0462	5.67 ^{q-x}	105.82 ^{j-l}	18.60 ^{t-r}	51.67 ^{b-e}	130.13 ^t	27.73 ^{f-k}	4.38 ^{t-h}	29.27 ^{n-u}	122.16 ^{b-b}	113.61 ^{r-i}	82.61 ^{o-t}
37	BC-0488	5.87 ^{o-v}	91.23 ^{p-s}	19.13 ^{e-p}	50.87 ^{c-i}	127.47 ^{a-a}	26.27 ^{g-o}	4.37 ^{u-h}	29.40 ^{m-u}	121.21 ^{v-d}	113.09 ^{y-j}	84.52 ^{h-s}
38	BC-0490	5.47 ^{s-y}	113.00 ^{b-i}	18.80 ^{h-q}	47.20 ^{t-y}	130.27 ^{k-t}	26.73 ^{g-n}	4.02 ^{d-l}	31.93 ^{d-o}	121.95 ^{o-c}	112.72 ^{c-j}	85.23 ^{c-r}
39	BC-0491	7.00 ^{g-l}	115.00 ^{a-g}	17.13 ^{q-t}	50.00 ^{f-l}	126.00 ^{z-b}	26.00 ^{h-q}	4.63 ^{l-b}	34.00 ^{b-e}	120.29 ^{a-d}	111.73 ^{i-k}	86.86 ^{c-m}
40	BC-0495	6.93 ^{g-m}	107.84 ^{h-k}	19.80 ^{d-k}	52.73 ^{ab}	130.93 ^{i-q}	25.13 ^{j-v}	5.23 ^{e-k}	31.13 ^{e-r}	121.43 ^{u-d}	112.69 ^{d-j}	88.44 ^{b-e}
41	BC-0509	6.07 ^{n-t}	124.83 ^{v-x}	19.93 ^{d-k}	49.53 ^{g-o}	129.00 ^{a-y}	23.67 ^{n-a}	4.64 ^{l-b}	31.67 ^{d-q}	122.78 ^{g-x}	114.06 ^{p-g}	85.82 ^{c-p}
42	BC-0510	9.00 ^{ab}	95.34 ^{o-q}	19.33 ^{d-o}	48.67 ^{l-s}	128.00 ^{a-a}	28.00 ^{e-j}	3.97 ^{g-l}	30.00 ^{j-t}	122.92 ^{e-x}	115.51 ^{c-s}	84.88 ^{d-s}
43	BC-0511	5.93 ^{o-u}	113.53 ^{b-i}	17.93 ^{m-s}	48.97 ^{k-q}	127.47 ^{a-a}	26.60 ^{g-n}	4.85 ^{j-u}	29.27 ^{n-u}	122.07 ^{n-b}	114.75 ^{j-b}	84.68 ^{f-s}
44	BC-0512	5.67 ^{q-x}	97.87 ^{m-p}	20.00 ^{d-j}	48.33 ^{m-t}	133.33 ⁱ	25.00 ^{j-v}	4.93 ^{h-r}	26.00 ^{vw}	123.16 ^{d-w}	113.54 ^{s-i}	87.30 ^{c-l}
45	BC-0513	4.80 ^{yz}	89.98 ^{q-s}	18.93 ^{g-p}	49.33 ^{j-o}	129.40 ^{a-x}	24.33 ^{l-x}	4.75 ^{k-y}	29.73 ^{k-u}	123.57 ^{c-s}	116.56 ^{a-k}	84.84 ^{d-s}
46	BC-0514	6.53 ^{i-p}	107.55 ^{i-k}	19.07 ^{e-p}	46.60 ^{u-a}	127.47 ^{a-a}	26.53 ^{g-n}	4.40 ^{t-h}	31.93 ^{d-o}	123.77 ^{c-p}	115.30 ^{f-v}	83.37 ^{m-t}
47	BC-0515	5.87 ^{o-v}	94.86 ^{o-r}	19.53 ^{d-o}	46.53 ^{v-a}	130.47 ^{k-r}	22.87 ^{q-b}	4.56 ^{o-c}	30.20 ^{i-t}	122.21 ^{n-a}	112.10 ^{g-k}	84.02 ^{k-s}
48	JA-11/N	4.87 ^{x-z}	108.63 ^{g-k}	19.20 ^{e-p}	49.73 ^{g-m}	127.67 ^{u-a}	22.53 ^{s-c}	4.57 ^{n-b}	31.33 ^{d-r}	123.04 ^{d-w}	114.29 ^{o-f}	88.14 ^{b-h}
49	JA-13/R	6.93 ^{g-m}	94.14 ^{o-r}	19.27 ^{d-o}	51.87 ^{b-d}	130.93 ^{i-q}	22.33 ^{t-c}	4.43 ^{s-g}	30.53 ^{h-t}	122.08 ^{b-b}	113.96 ^{p-g}	81.38 ^{s-u}
50	JA-08/B	5.07 ^{v-z}	94.13 ^{o-r}	19.63 ^{d-n}	51.00 ^{c-g}	130.60 ^{l-r}	25.87 ^{h-r}	4.68 ^{l-z}	30.60 ^{g-t}	122.22 ^{b-a}	114.34 ^{o-f}	85.34 ^{c-r}
51	JA-11/L	7.00 ^{g-l}	167.79 ^{bc}	17.00 ^{r-t}	46.67 ^{u-a}	127.00 ^{b-b}	27.33 ^{f-l}	4.76 ^{k-y}	29.67 ^{l-u}	128.54 ^a	120.50 ^a	87.78 ^{c-j}
52	JA-17/2	7.00 ^{g-l}	148.79 ^{i-m}	20.00 ^{d-j}	46.67 ^{u-a}	134.67 ^{f-f}	27.33 ^{f-l}	4.29 ^{y-i}	31.00 ^{f-s}	124.59 ^{a-j}	117.46 ^{a-d}	87.78 ^{c-j}
53	JA-13/X	7.00 ^{g-l}	151.35 ^{g-k}	19.73 ^{d-l}	50.00 ^{f-l}	126.00 ^{z-b}	27.00 ^{f-m}	4.46 ^{r-f}	30.00 ^{j-t}	124.97 ^{a-e}	116.62 ^{a-j}	81.62 ^{t-u}
54	JA-09/G	4.53 ^z	150.20 ^{j-l}	17.87 ^{n-s}	50.20 ^{e-l}	128.67 ^{q-y}	28.00 ^{e-j}	4.60 ^{l-b}	29.67 ^{l-u}	120.94 ^{c-d}	112.84 ^{b-j}	81.33 ^{s-u}
55	JA-08/4	5.07 ^{v-z}	153.30 ^{f-j}	18.53 ^{t-r}	49.60 ^{g-n}	130.47 ^{k-r}	25.73 ^{i-r}	4.82 ^{k-v}	30.40 ^{h-t}	122.54 ^{i-y}	114.67 ^{d-j}	88.45 ^{b-e}
56	JA-085	6.60 ^{h-o}	157.77 ^{d-g}	19.40 ^{d-o}	50.20 ^{e-l}	130.93 ^{i-q}	24.80 ^{k-w}	4.37 ^{u-h}	29.00 ^{o-u}	122.17 ^{n-b}	114.21 ^{o-f}	87.94 ^{b-i}
57	JA-0510	7.07 ^{g-k}	157.76 ^{d-g}	20.07 ^{d-i}	47.13 ^{s-z}	131.93 ⁿ	25.47 ^{j-t}	4.98 ^{h-q}	31.73 ^{d-p}	121.13 ^{w-d}	113.89 ^{p-h}	85.34 ^{c-r}
58	Mutant-1	8.13 ^{c-e}	136.08 ^{q-s}	20.13 ^{d-i}	47.00 ^{t-z}	127.33 ^{w-b}	27.00 ^{f-m}	4.42 ^{t-g}	28.00 ^{t-v}	121.11 ^{w-d}	112.11 ^{g-k}	86.45 ^{c-n}

Table 8. Mean values based on phenotypic expressions of 11 morphological characteristics in 100 upland cotton genotypes

SL.	Genotype	PA	PH (cm)	SBPP	DFF	DFBS	BPP	SBW (g)	SPB	SCYPP (g)	LFYPP (g)	EI (%)
59	TC-1901	5.20 ^{u-z}	135.50 ^{q-s}	19.60 ^{d-n}	45.40 ^{a-c}	129.80 ^{l-v}	24.47 ^{l-x}	4.45 ^{r-g}	31.73 ^{d-p}	123.37 ^{d-u}	114.56 ^{k-d}	87.37 ^{c-k}
60	TC-1902	5.20 ^{u-z}	141.63 ^{n-q}	19.07 ^{e-p}	49.33 ^{i-o}	129.80 ^{l-v}	27.00 ^{f-m}	4.36 ^{v-h}	32.73 ^{c-j}	121.25 ^{v-d}	114.20 ^{o-f}	83.55 ^{l-s}
61	TC-1903	8.33 ^{b-d}	146.03 ^{k-o}	21.00 ^{b-d}	48.33 ^{m-t}	135.00 ^{c-f}	31.00 ^{b-e}	4.01 ^{e-l}	31.00 ^{f-s}	124.78 ^{a-g}	115.77 ^{b-p}	84.88 ^{d-s}
62	TC-1904	5.00 ^{w-z}	158.08 ^{d-f}	18.00 ^{l-s}	47.33 ^{r-x}	133.00 ^{c-f}	25.00 ^{j-v}	5.30 ^{e-j}	32.00 ^{d-n}	124.45 ^{a-k}	115.50 ^{d-t}	86.45 ^{c-n}
63	BC-0024	7.20 ^{f-j}	158.22 ^{d-f}	19.00 ^{f-p}	44.33 ^c	128.00 ^{s-a}	25.67 ^{i-s}	4.16 ^{b-k}	31.00 ^{f-s}	120.68 ^{v-d}	113.17 ^{x-j}	93.00 ^a
64	BC-0025	7.00 ^{g-l}	175.34 ^a	21.00 ^{b-d}	49.67 ^{g-n}	130.00 ^{l-u}	29.00 ^{d-h}	4.35 ^{v-h}	30.00 ^{j-t}	126.25 ^a	116.81 ^{a-i}	84.88 ^{d-s}
65	BC-0026	5.00 ^{w-z}	152.71 ^{f-j}	19.00 ^{f-p}	46.33 ^{w-b}	132.00 ^{g-m}	24.33 ^{l-x}	5.41 ^{d-h}	28.00 ^{t-v}	124.36 ^{a-l}	114.48 ^{l-e}	87.78 ^{c-j}
66	BC-0027	8.67 ^{a-c}	137.07 ^{p-s}	20.00 ^{d-j}	44.67 ^c	136.00 ^{b-d}	32.00 ^{a-d}	3.79 ^{j-l}	33.00 ^{c-i}	121.22 ^{v-d}	113.50 ^{s-i}	91.65 ^{ab}
67	BC-0028	7.67 ^{d-g}	123.92 ^{v-x}	16.33 st	49.33 ^{i-o}	127.00 ^{x-b}	27.00 ^{f-m}	4.52 ^{b-c}	37.00 ^a	123.64 ^{c-r}	116.56 ^{a-k}	86.45 ^{c-n}
68	BC-0029	6.47 ^{j-q}	158.04 ^{d-f}	20.00 ^{d-j}	48.33 ^{m-t}	129.00 ^{o-y}	29.00 ^{d-h}	4.30 ^{y-i}	30.00 ^{j-t}	124.76 ^{a-g}	117.11 ^{a-h}	86.45 ^{c-n}
69	BC-0030	6.40 ^{j-r}	143.01 ^{m-p}	18.27 ^{j-r}	47.47 ^{q-w}	128.73 ^{q-y}	29.27 ^{d-g}	3.92 ^{h-l}	31.80 ^{d-p}	121.57 ^{s-d}	113.50 ^{s-i}	85.78 ^{c-p}
70	BC-0031	5.73 ^{p-w}	134.17 ^{r-u}	17.87 ^{n-s}	50.93 ^{c-h}	132.60 ^{f-k}	21.40 ^{x-c}	4.66 ^{l-a}	29.87 ^{j-u}	122.60 ^{j-y}	115.40 ^{e-u}	81.87 ^{q-u}
71	BC-0032	5.60 ^{r-y}	123.98 ^{v-x}	20.67 ^{b-g}	46.33 ^{w-b}	129.67 ^{m-w}	28.93 ^{d-h}	4.93 ^j	33.27 ^{b-h}	123.21 ^{d-v}	114.37 ^{n-f}	87.41 ^{c-k}
72	BC-0033	8.67 ^{a-c}	117.07 ^{y-d}	18.00 ^{l-s}	47.00 ^{t-z}	140.00 ^a	34.33 ^a	4.28 ^{y-i}	31.00 ^{f-s}	126.10 ^{ab}	116.36 ^{b-n}	88.33 ^{b-f}
73	BC-0035	6.13 ^{m-t}	102.66 ^{k-m}	19.87 ^{d-k}	45.80 ^{x-c}	131.33 ^{b-p}	26.13 ^{g-p}	3.60 ^l	32.00 ^{d-n}	122.96 ^{d-x}	115.33 ^{f-v}	84.64 ^{f-s}
74	BC-0036	9.00 ^{ab}	123.54 ^{v-z}	16.00 ^l	47.00 ^{t-z}	138.00 ^{ab}	31.13 ^{b-e}	4.16 ^{b-k}	29.00 ^{o-u}	123.90 ^{c-o}	114.92 ^{a-i}	87.78 ^{c-j}
75	BC-0037	8.00 ^{c-f}	114.45 ^{a-h}	17.00 ^{r-t}	47.00 ^{t-z}	134.00 ^{d-g}	29.00 ^{d-h}	4.24 ^{z-j}	31.00 ^{f-s}	125.43 ^{a-c}	117.53 ^{a-c}	87.78 ^{c-j}
76	BC-0038	6.50 ^{i-q}	115.46 ^{a-f}	16.33 st	47.00 ^{t-z}	128.00 ^{s-a}	24.00 ^{m-z}	5.31 ^{e-j}	22.00 ^x	124.69 ^{a-h}	116.50 ^{a-l}	87.78 ^{c-j}
77	BC-0039	5.07 ^{v-z}	116.26 ^{a-e}	18.93 ^{g-p}	45.27 ^{a-c}	129.33 ^{o-x}	27.47 ^{f-l}	4.99 ^{g-p}	32.00 ^{d-n}	122.57 ^{j-y}	113.74 ^{q-l}	82.52 ^t
78	BC-0040	6.20 ^{l-s}	102.77 ^{k-m}	20.67 ^{b-g}	47.73 ^{p-w}	131.00 ^{l-q}	15.80 ^g	4.62 ^{l-b}	31.87 ^{d-o}	122.89 ^{f-x}	115.60 ^{b-r}	85.06 ^{c-s}
79	BC-0041	4.60 ^z	95.94 ^{a-q}	18.20 ^{k-r}	47.20 ^{r-y}	131.07 ^{i-q}	16.87 ^{ef}	4.81 ^{k-w}	30.00 ^{j-t}	122.21 ^{u-a}	115.10 ^{h-y}	86.32 ^{c-o}
80	BC-0042	3.33 ^a	135.77 ^{q-s}	18.00 ^{l-s}	47.33 ^{r-x}	130.67 ^{j-r}	13.00 ^g	6.25 ^{ab}	25.00 ^w	112.58 ^f	101.58 ^m	86.45 ^{c-n}
81	BC-0043	4.87 ^{x-z}	88.29 ^{is}	19.13 ^{e-p}	44.33 ^c	127.87 ^{t-a}	20.87 ^{z-d}	4.66 ^{l-a}	32.27 ^{c-m}	121.47 ^{t-d}	113.49 ^{i-k}	84.17 ^s
82	BC-0044	5.60 ^{r-y}	123.70 ^{v-y}	19.07 ^{e-p}	49.40 ^{h-o}	133.00 ^{e-j}	22.73 ^c	4.49 ^{r-e}	28.73 ^{q-v}	122.62 ^{j-y}	116.12 ^{b-o}	84.51 ^{h-s}
83	BC-0045	5.00 ^{w-z}	131.52 ^{s-u}	20.00 ^{d-j}	47.33 ^{r-x}	135.33 ^{c-e}	22.00 ^{v-c}	5.33 ^{e-j}	29.00 ^{o-u}	117.35 ^e	110.50 ^{kl}	86.45 ^{c-n}
84	BC-0046	5.00 ^{w-z}	133.08 ^{s-u}	19.00 ^{f-p}	47.00 ^{t-z}	128.67 ^{q-y}	24.00 ^{m-z}	4.50 ^{g-p}	29.00 ^{o-u}	120.00 ^d	112.96 ^{a-j}	87.78 ^{c-j}
85	BC-0047	5.00 ^{w-z}	127.54 ^{u-w}	19.00 ^{f-p}	49.33 ^{i-o}	126.00 ^{z-b}	25.00 ^{j-v}	4.79 ^{k-x}	31.00 ^{f-s}	119.85 ^d	111.73 ^{k-i}	86.45 ^{c-n}
86	BC-0048	5.00 ^{w-z}	128.20 ^{t-v}	19.67 ^{d-m}	49.33 ^{i-o}	133.33 ^{e-i}	21.00 ^{y-d}	6.12 ^{a-c}	27.00 ^{u-w}	122.33 ^{l-a}	115.09 ^{y-i}	81.62 ^{r-u}
87	BC-0049	3.00 ^a	112.39 ^{c-j}	18.00 ^{l-s}	49.33 ^{i-o}	135.33 ^{c-e}	18.00 ^{d-f}	6.44 ^a	32.00 ^{d-n}	116.23 ^e	108.54 ^l	86.45 ^{c-n}
88	BC-0050	7.00 ^{g-l}	134.73 ^{r-t}	17.00 ^{r-t}	49.00 ^{k-q}	130.00 ^{l-u}	26.00 ^{b-q}	4.76 ^{k-y}	28.00 ^{t-v}	123.64 ^{c-r}	114.86 ^{i-b}	84.88 ^{d-s}
89	BC-0051	4.80 ^{yz}	142.07 ^{n-q}	19.53 ^{d-o}	50.53 ^{d-k}	133.73 ^{d-h}	25.53 ^{l-s}	5.08 ^{f-l}	30.80 ^{t-t}	122.81 ^{g-x}	116.21 ^{b-o}	85.37 ^{c-q}
90	BC-0052	8.67 ^{a-c}	131.70 ^{s-u}	16.00 ^l	50.00 ^{f-l}	130.00 ^{l-u}	31.00 ^{b-e}	3.98 ^{f-l}	28.00 ^{t-v}	123.24 ^{d-v}	117.13 ^{a-g}	81.62 ^{r-u}
91	BC-0053	5.00 ^{w-z}	149.59 ^{j-m}	20.00 ^{d-j}	47.33 ^{r-x}	138.00 ^{ab}	24.00 ^{m-z}	5.17 ^{e-k}	27.00 ^{u-w}	124.07 ^{b-n}	115.74 ^{b-q}	91.65 ^{ab}
92	BC-0054	7.33 ^{e-i}	135.53 ^{q-s}	22.20 ^{ab}	51.33 ^{b-f}	136.67 ^{bc}	27.00 ^{f-m}	4.59 ^{m-b}	33.00 ^{c-i}	124.67 ^{a-i}	117.61 ^{ab}	81.62 ^{r-u}
93	BC-0055	8.33 ^{b-d}	134.45 ^{r-t}	18.00 ^{l-s}	49.00 ^{k-q}	129.00 ^{o-y}	33.00 ^{a-c}	3.71 ^{kl}	27.00 ^{u-w}	122.34 ^{l-a}	114.61 ^{j-d}	84.88 ^{d-s}
94	BC-0056	5.67 ^{q-x}	153.53 ^{e-j}	20.07 ^{d-i}	49.33 ^{i-o}	134.80 ^{c-f}	21.80 ^{w-c}	4.29 ^{y-i}	30.80 ^{t-t}	122.74 ^{g-x}	116.21 ^{b-o}	88.65 ^{bc}
95	BC-0057	5.67 ^{q-x}	156.90 ^{d-h}	23.18 ^a	49.00 ^{k-q}	128.00 ^{s-a}	22.00 ^{v-c}	5.51 ^{d-f}	28.00 ^{t-v}	121.23 ^{v-d}	113.08 ^{y-j}	86.45 ^{c-n}
96	BC-0058	7.00 ^{g-l}	172.89 ^{ab}	19.67 ^{d-m}	49.00 ^{k-q}	126.00 ^{z-b}	26.00 ^{b-q}	4.62 ^{l-b}	31.00 ^{f-s}	120.13 ^{b-d}	116.19 ^{b-o}	84.88 ^{d-s}
97	BC-0059	5.00 ^{w-z}	143.31 ^{m-p}	21.00 ^{b-d}	49.00 ^{k-q}	134.00 ^{d-g}	24.00 ^{m-z}	5.05 ^{f-n}	34.00 ^{b-e}	121.21 ^{v-d}	117.41 ^{a-e}	84.88 ^{d-s}
98	BC-0060	7.00 ^{g-l}	119.31 ^{x-b}	20.00 ^{d-j}	48.67 ^{l-s}	130.00 ^{l-u}	29.00 ^{d-h}	4.31 ^{x-i}	29.00 ^{o-u}	125.00 ^{a-d}	116.09 ^{b-o}	84.88 ^{d-s}
99	BC-0061	6.33 ^{k-r}	118.96 ^{x-c}	20.00 ^{d-j}	54.00 ^a	128.00 ^{s-a}	29.00 ^{d-h}	4.19 ^{a-k}	30.00 ^{j-t}	121.43 ^{u-d}	114.37 ^{n-f}	76.21 ^v
100	BC-0062	9.20 ^a	168.48 ^{bc}	22.00 ^{a-c}	54.00 ^a	132.00 ^{g-m}	34.00 ^{ab}	3.63 ^l	28.87 ^{p-v}	123.33 ^{d-u}	116.41 ^{b-m}	77.09 ^v
SD		1.27	25.18	1.30	2.17	2.95	3.70	0.57	2.23	1.96	2.31	2.77
LSD (5%)		0.86	6.67	1.78	1.58	2.44	3.19	0.48	2.98	2.06	2.02	3.75

Here, same letter in superscript indicates no significant difference, $p < 0.05$, PA= phenotypic acceptability, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, DFBS= days to first boll splitting, BPP= bolls per plant, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness index (%), SD= standard deviation and LSD (5%)= least significant difference at 5% level of probability.

Single boll weight varied from 3.60-6.44 g in the current study. The genotype CB-8, BC-0049, BC-0042, CB-6, BC-0048 had > 6 g of single boll weight. Boll weight is a crucial

component that has a direct impact on yield. The increased yield was related to a greater quantity of bolls per plant as well as a greater quantity of single boll weight. Mawblei *et al.*, (2022) reported Acala-1577-D (4.67 g) showed the highest performance for boll weight. Rehman *et al.*, (2020) reported that the genotype VH-367 scored highest mean estimate of 2.46 g whereas, CIM-632 had least mean estimate for boll weight of 1.83g.

In the present study, seeds per boll varied from 22-37. The genotype BC-0028 had the highest number of seeds per boll of 37. Rehman *et al.*, (2020) reported that for 100-seed weight, the accessions NIAB-414 and A555 demonstrated the lowest and the highest mean estimations of 5.07 and 5.64 g, respectively.

Among the 100 upland cotton genotypes, JA-11/L exhibited the greatest seed cotton yield per plant, measuring 128.54g, followed by BC-0025 and BC-0033 which achieved seed cotton yield per plant of 126.25g and 126.10g, respectively (**Table 8**). The genotype BC-0042 had the lowest number of seed cotton yield per plant of 112.58g followed by BC-0049 and BC-0045 of 116.23 and 117.35g per plant. Rehman *et al.*, (2020) opined that IUB-222 scored the highest mean value of 74.81g for seed cotton yield. Mawblei *et al.*, (2022) reported that 16 genotypes exhibited elevated yield based on the mean performance among 100 upland cotton germplasms. Mahdy *et al.*, (2023) showed that mean seed cotton yield per plant of the parents Giza90 and Giza86 was 104.67 and 94.06g under normal field condition.

Lint fiber yield per plant varied between 101.58-120 g per plant. The genotype JA-11/L had the highest lint fiber yield per plant of 120g whereas the genotype BC-0042 showed the lowest lint fiber yield per plant of 101.58 g. Hence, these can serve as a functional donor for forthcoming breeding programs in cotton. Mahdy *et al.*, (2018) showed that the reduction in lint yield was more than that in seed cotton yield.

Earliness index varied between 76.21 % - 94 %. The genotype BC-0024, RA-2, BC-0027, BC-0053 are the early maturing genotypes, showed maximum values for earliness index of 93 %, and 91.65 %. The genotype BC-0061, BC-0062, CB-2, CB-5 were the late maturing genotypes which exhibited earliness index of < 80 %. Mahdy *et al.*, (2018) opined that G. 90 flowered normal and showed the highest earliness index, indicating that G. 90 flowered normal and gave most of its yield in few weeks. Amer & Hassan, (2023) examined that earliness index was 58.41% for Giza 87, which indicated more delay maturity to 82.39% for promising line No.3, the more earliness of this genotype

might be attributed to superior genetic makeup compared to Giza 87. Abubakar *et al.* (2024) showed that the greater earliness index was recorded in PB-130 (67.06) and the lowest earliness index was recorded in PB-94 (42.73) when analyzed 14 genotypes of cotton.

4.1.3 Variability study among the 100 upland cotton genotypes

The ability to improve economic characteristics via genotype selection is primarily contingent upon the degree of genetic variability. The genotypic and phenotypic coefficients of variation reflect the degree of genetic variability within a population, but heritability facilitates the prediction of the impact of transmission factors on phenotype expression in succeeding generations, thereby informing the process of selecting better genotypes through advancing generations. Heritability with genetic advance, guarantees the stability of genetic parameters for concerning polygenic traits.

The outcomes of several genetic parameters for eleven traits are presented in **Table 9**. The findings indicated that the observed variations in phenotypic (σ^2_p) traits were greater than the variations in genetic (σ^2_g) makeup for all the characteristics, suggesting strong influence of environment upon expression of the traits.

High σ^2_g and σ^2_p were recorded with plant height (628.23 and 645.39) followed by bolls per plant (12.37 and 16.30), days to first boll splitting (7.93 and 10.22) and low values which ranged from >1 to 2 were observed with the character single boll weight (0.30 and 0.39) followed by sympodial branches per plant (1.29 and 2.51) and phenotypic acceptability (1.53 and 1.82), respectively.

The phenotypic coefficient of variation (PCV) was higher than genotypic coefficient of variation (GCV) for all the traits. Based on Burton & De Vane (1953) classification of PCV and GCV, the current study revealed elevated values of GCV and PCV (20.06 % and 21.84 %) for phenotypic acceptability attributed to significant heterogeneity among the genotypes examined for the attributes. The similarity between the two values indicated very little environmental influence on these traits, highlighting a significant potential for enhancement through selection. The results aligned with Pandiyan *et al.*, 2019; Praveen *et al.*, 2019; Reddy *et al.*, 2019; Aarthi *et al.*, 2018.

Table 9. Genetic parameters of 11 morphological characteristics in 100 upland cotton genotypes

Character	σ^2_g	σ^2_p	GCV	PCV	h^2_b (%)	GA	GAM
PA	1.53	1.82	20.06	21.84	84.35	2.34	37.95
PH	628.23	645.39	18.89	19.15	97.34	50.94	38.39
SBPP	1.29	2.51	5.92	8.27	51.31	1.68	8.74
DFF	4.38	5.33	4.31	4.76	82.07	3.91	8.05
DFBS	7.93	10.22	2.16	2.45	77.56	5.11	3.92
BPP	12.37	16.30	13.70	15.73	75.92	6.31	24.59
SBW	0.30	0.39	11.66	13.31	76.79	0.99	21.06
SPB	3.82	7.23	6.36	8.76	52.78	2.92	9.52
SCYPP	3.30	4.94	1.48	1.81	66.76	3.06	2.49
LFYPP	4.79	6.36	1.91	2.20	75.26	3.91	3.42
EI	5.85	11.27	2.83	3.92	51.90	3.59	4.20

Here, PA= phenotypic acceptability, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, DFBS= days to first boll splitting, BPP= bolls per plant, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness index (%), σ^2_g = genotypic variance, σ^2_p = phenotypic variance, GCV= genotypic coefficient of variance, PCV= phenotypic coefficient of variance, h^2_b = heritability in broad sense (%), GA= genetic advance, and GAM= genetic advance as percentage of mean.

Average GCV and PCV were assessed for plant height (18.89 % and 19.15 %), bolls per plant (13.70 % and 15.73 %) and single boll weight (11.66 % and 13.31 %) indicating the existence of a moderate degree of magnitudes that can be utilized through selection for an effective breeding effort. The results validated the outcomes of Pandiyan *et al.*, 2019; Shruti *et al.*, 2019; Aarthi *et al.*, 2018; Dhivya *et al.*, 2014.

Low GCV and PCV were accounted for the traits like seeds per boll (6.36 % and 8.76 %), sympodial branches per plant (5.92 % and 8.27 %), days to 50 % flowering (4.31 % and 4.76 %), earliness index (2.83 % and 9.92 %), days to first boll splitting (2.016 % and 2.45 %), lint fiber yield per plant (1.91 % and 2.20 %) and seed cotton yield per plant (1.48 % and 1.81 %) demonstrating reduced variability among the examined genotypes (**Table 9**). Comparable outcomes were also suggested by Pandiyan *et al.*, 2019; Reddy *et al.*, 2019; Shruti *et al.*, 2019; Aarthi *et al.*, 2018.

In this study, the heritability (h^2_b) estimation ranged from 51.31 % to 97.34 %. Plant height (97.34 %), phenotypic acceptability (84.35 %), days to 50 % flowering (82.07 %), days to first boll splitting (77.56 %), single boll weight (76.79 %), bolls per plant (75.92 %), lint fiber yield per plant (75.26 %) and seed cotton yield per plant (66.76 %) demonstrated high h^2_b values as per the categories described by Robinson *et al.*, 1949, Falconer & Mackay, 1996. This recommended that additive gene action primarily influenced above traits and, consequently, can be effectively targeted for selection and improvement in future breeding programs. It was aligned to prior findings of Patel *et al.*, 2023; Deshmukh *et al.*, 2019; Manonmani *et al.*, 2019; Pandiyan *et al.*, 2019; Praveen *et al.*, 2019; Reddy *et al.*, 2019; Shruti *et al.*, 2019.

Heritability estimates alone are less helpful than heritability coupled with genetic advances in projecting yield under phenotypic selection, according to earlier research of Johnson *et al.*, 1955; Swarup & Chaugale, 1962. When heritability is highly influenced by non-additive like dominant and epistatic gene effects the genetic advance tends to be low. Conversely, if a significant additive gene effect is present, a higher genetic advance is anticipated (Panse & Sukhatme, 1978; Panse, 1957).

The most accurate measure of the expected progress resulting from the selection can be derived from the integration of heritability, genotypic coefficient of variation, and genetic advance (Johnson *et al.*, 1955). The genetic advance as percentage of mean (GAM) varied from 2.49 % for seed cotton yield per plant to 38.39% for plant height

(**Table 9**). Out of eleven traits, four traits such as plant height (38.39 %), phenotypic acceptability (37.95 %), bolls per plant (24.59 %) and single boll weight (21.06 %) exhibited elevated levels of GAM (>20 %) coupled with high heritability in accordance with Falconer & Mackay, 1996; Johnson *et al.*, 1955. This suggested that heritability was predominantly responsible to additive genetic effects, thus, selection may be advantageous by exploiting the heritable genes for enhancement. Meena *et al.*, (2022a) indicated that monopodia per plant (39.26 %), sympodia per plant (22.50 %), plant height (21.39 %), bolls per plant (58.58 %), seed cotton yield per plant (52.97 %) and lint yield per plant (56.39 %) exhibited high h^2_b along with high GAM. Comparable outcomes were also shown by Pandiyan *et al.*, 2019; Praveen *et al.*, 2019; Aarthi *et al.*, 2018; Monisha *et al.*, 2018.

Elevated heritability alongside minimal GAM was estimated for days to 50% flowering (8.05 %), days to first boll splitting (3.92 %), lint fiber yield per plant (3.42 %), and seed cotton yield per plant (2.49 %), suggesting the existence of non-additive gene action that restricts potential for enhancement. Meena *et al.*, (2022a) observed significant heritability alongside minimal GAM for days to flowering (8.45 %) and oil content (5.88 %). Manonmani *et al.*, 2019; Eswari *et al.*, 2017; Erande *et al.*, 2014 obtained analogous results.

Moderate heritability with a low GAM, was seen for seeds per boll (9.52), sympodial branches per plant (8.74), and earliness index (4.20 %), signifying the prevalence of non-additive genetic influence (**Table 9**). Heritability was shown as a result of strong genotype x environment interaction rather than genotype itself, suggesting that this trait may be enhanced by recurrent selection instead of simple selection. Meena *et al.*, (2022a) indicated that days to boll bursting (4.70 %), micronaire (7.06 %) and 2.5 % span length (3.46 %) exhibited average heritability with poor GAM. Praveen *et al.*, 2019; Monisha *et al.*, 2018; Erande *et al.*, 2014 reported analogous findings.

4.1.4 Correlation coefficient among the 100 upland cotton genotypes

The correlation coefficient, denoted as “r”, provides information regarding the extent and order of the linear relation (Al-Jibouri *et al.*, 1958). The present study examined the relationships in both phenotypic and genotypic levels among eleven variables. The genotypic correlation demonstrates an intrinsic connection between genes that control two distinct traits, hence facilitating the implementation of an effective selection strategy. The phenotypic correlation is an unreliable indicator of the association between

two qualities since it is influenced by environmental factors. The genotypic and phenotypic correlation coefficients of eleven morphological characteristics in 100 upland cotton genotypes are evidenced in **Table 10**.

The parameter seed cotton yield per plant revealed a strong positive genotypic and phenotypic correlation with lint fiber yield per plant (0.820** and 0.776**), bolls per plant (0.469** and 0.297**) and phenotypic acceptability (0.442** and 0.326**). The strong correlations between these pairs of characteristics suggested that they had a substantial impact on successful selection for increased productivity. Similar attributes were also observed by Patel *et al.*, 2023; Chapepa *et al.*, 2020; Nawaz *et al.*, 2019; Gnanasekaran *et al.*, 2018. Mahdy *et al.* (2023) examined that the correlations of lint yield per plant behaved the same as seed cotton yield per plant. Plant height (0.031 and 0.017) and days to first boll splitting (0.068 and 0.051) showed positive genotypic and phenotypic correlations to seed cotton yield per plant. Patel *et al.*, (2023) reported that traits such as boll weight, seed index, lint index, seed oil content, fiber fineness and fiber strength, exhibited non-significant associations with seed cotton yield. Relatable outcomes were also attained by Rai & Sangwan, 2020; Nawaz *et al.*, 2019; Nikhil *et al.*, 2018.

On the other hand, the parameter sympodial branches per plant (-0.026 and -0.043), days to 50% flowering (-0.022 and -0.243), single boll weight (-0.406** and -0.278**), seeds per boll (-0.019 and -0.003) and earliness index (-0.048 and -0.007) displayed strong negative genotypic and phenotypic correlation with seed cotton yield per plant (**Table 10**). The strong association between seed cotton yield and these traits can not be advantageously utilized in the selection program to cultivate high-yielding genotypes, might be due to strong genotype x environment interaction as well as imbalanced physiological pathways in reproductive phase of cotton plants. These assumptions were aligned with the finding reported by Rehman & Farooq (2019). Patel *et al.*, (2023) showed that there were strong negative associations between seed cotton yield and plant height, the number of monopodial branches per plant, the days to 50% boll bursting, and ginning outturn.

Phenotypic acceptability demonstrated strong positive genotypic and phenotypic correlation with bolls per plant (0.752** and 0.609**), seed cotton yield per plant (0.442** and 0.326**) and lint fiber yield per plant (0.384** and 0.302**) but strong negative correlation with single boll weight (-0.668** and -0.533**).

Table 10. Genotypic and phenotypic correlation coefficients of 11 morphological characteristics in 100 upland cotton genotypes

Character	Correlation co-efficient	PH	SBPP	DFP	DFBS	BPP	SBW	SPB	SCYPP	LFYPP	EI
PA	r _g	0.073	-0.066	0.001	0.083	0.752**	-0.668**	-0.015	0.442**	0.384**	-0.029
	r _p	0.065	-0.045	0.001	0.064	0.609**	-0.533**	-0.033	0.326**	0.302**	0.001
PH	r _g		0.271**	0.011	0.035	0.180	0.172	0.130	0.031	-0.000	0.023
	r _p		0.205**	0.015	0.023	0.149**	0.148*	0.087	0.017	-0.002	0.017
SBPP	r _g			0.103	0.067	-0.107	-0.002	0.086	-0.026	-0.010	-0.145
	r _p			0.068	0.049	-0.023	0.031	0.047	-0.043	0.014	-0.130*
DFP	r _g				-0.056	0.131	0.029	-0.163	-0.022	0.129	-0.565**
	r _p				-0.035	0.090	0.013	-0.099	-0.243	0.101	-0.024
DFBS	r _g					0.063	-0.067	-0.026	0.068	0.118	0.245*
	r _p					0.034	-0.019	-0.015	0.051	0.094	0.175**
BPP	r _g						-0.576**	0.100	0.469**	0.398**	-0.131
	r _p						-0.464**	0.044	0.297**	0.282**	-0.048
SBW	r _g							-0.058	-0.406**	-0.427**	0.085
	r _p							-0.020	-0.278**	-0.321**	0.040
SPB	r _g								-0.019	0.073	0.106
	r _p								-0.003	0.050	0.075
SCYPP	r _g									0.820**	-0.048
	r _p									0.776**	-0.007
LFYPP	r _g										-0.057
	r _p										-0.038

Here, * and ** indicated significant at 5 % and 1 % and levels of probability, respectively; PA= phenotypic acceptability, PH= plant height (cm), SBPP= sympodial branches per plant, DFP= days to 50% flowering, DFBS= days to first boll splitting, BPP= bolls per plant, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness index (%), r_g= correlation coefficient at genotypic level and r_p= correlation coefficient at phenotypic level.

Plant height showed notable and meaningful positive genotypic and phenotypic correlations with sympodial branches per plant (0.271** and 0.205**), but significant positive phenotypic correlation with bolls per plant (0.180 and 0.149**) and single boll weight (0.172 and 0.148*) (**Table 10**). This correlation is logical and increasing number of sympodial branches per plant with plant height might produce higher seed cotton yield. These results confirmed the investigation carried out by Patel *et al.*, 2023.

Therefore, selecting based on phenotypic acceptability, bolls per plant, sympodial branches per plant, in conjunction with boll weight and lint yield per plant, could lead to a significant breakthrough in enhancing seed cotton yield. Similar results were reported in studies carried out by Rao & Gopinath, 2013; Kalpande *et al.*, 2008.

4.1.5 Path analysis among the 100 upland cotton genotypes

Path coefficient analysis is an important biometrical analysis which measures direct and indirect contributions of various attributes on yield per plant (Dewey & Lu, 1959). In agriculture, the analysis of path coefficients allows breeders focusing on traits that exhibit a significant direct effect on production, thereby aiding in the identification of features that serve as effective selection criteria for enhancing crop yield. Here, the path coefficient analysis was executed using correlation co-efficient to evaluate the direct and indirect influences of the eleven traits on seed cotton yield per plant both at genotypic and phenotypic levels (**Table 11**).

The genotypic path analysis disclosed that lint fiber yield per plant imposed highest direct positive effect (0.797) on seed cotton yield followed by bolls per plant (0.216), single boll weight (0.087), sympodial branches per plant (0.036) and phenotypic acceptability (0.033). Whereas, plant height (-0.013), days to first boll splitting (-0.034), earliness index (-0.083), seeds per boll (-0.124) and days to 50% flowering (-0.228) imposed negative direct effect on seed cotton yield per plant. Similar trend of direct associations was revealed at phenotypic path analysis. This implied that the direct selection of genotypes exhibiting these characteristics can successfully result in high-yielding genotypes. Kadam *et al.*, (2024) reported that plant height (0.510), bolls per plant (0.404), uniformity ratio (0.339), ginning outturn (0.323), fiber strength (0.080) and lint index (0.029) enforced direct positive effect on seed cotton yield per plant at genotypic level while, days to 50 percent flowering, number of sympodia per plant, seed index, upper half mean length and micronnaire value enforced negative direct effect on

seed cotton yield per plant. These results validated with the results of Shruti *et al.*, 2020; Chaudhari *et al.*, 2017; Farias *et al.*, 2016; Dahiphale *et al.*, 2015; Latif *et al.*, 2015)

Here, the phenotypic path coefficient showed that lint fiber yield per plant (0.765) had highest positive direct impact on seed cotton yield per plant followed by phenotypic acceptability (0.073), bolls per plant (0.063), single boll weight (0.036) and plant height (0.014). Therefore, direct selection for these traits is advised to increase yield in cotton. Mawblei *et al.*, (2022) reported that the number of bolls per plant (0.649), boll weight (0.299), plant height (0.058) and monopodia per plant (0.027) had elevated positive direct effects on the seed cotton yield, while, days to 50% flowering (0.004) showed slightly positive direct effect on seed cotton yield. Nawaz *et al.*, (2019) showed positive direct effects of bolls per plant, boll weight and plant height, but a negative direct effect on seed cotton yield by monopodia per plant.

In the present study, each attribute exhibited both positive and negative indirect impacts on the yield per plant at both levels. At genotypic level, the trait phenotypic acceptability exerted maximum positive indirect effect on seed cotton yield per plant via lint fiber yield per plant (0.306) and bolls per plant (0.163) but negative indirect effect through single boll weight (-0.058). The trait days to 50% flowering exerted positive indirect effect on seed cotton yield per plant via lint fiber yield per plant, earliness index, bolls per plant and seeds per boll (**Table 11**). Kadam *et al.*, (2024) opined that positive indirect effect on yield was exerted through boll weight (0.054), bolls per plant (0.038), seed index (0.036), sympodia per plant (0.025) and ginning outturn (0.013). These results were in agreement with Sainath *et al.*, 2022; Shruti *et al.*, 2020; Gulhane & Wadikar, 2017.

At phenotypic level, the traits bolls per plant displayed a positive indirect effect on seed cotton yield through lint fiber yield per plant (0.215), phenotypic acceptability (0.045) but a negative indirect effect through single boll weight (-0.017) and days to 50% flowering (-0.011).

Again, the trait days to 50% flowering had positive indirect effect on seed cotton yield per plant through lint fiber yield per plant (0.077) at phenotypic level. However, it was conflicting with the results of Kadam *et al.*, (2024) who found that days to 50% flowering had negative direct effect (-0.215) on yield.

Table 11. Genotypic and phenotypic path coefficients of 10 morphological characteristics to seed cotton yield per plant in 100 upland cotton genotypes

Character	Path co-efficients	PA	PH	SBPP	DFF	DFBS	BPP	SBW	SPB	LFYPP	EI	SCYPP
PA	G	0.033	-0.001	-0.002	0.000	-0.003	0.163	-0.058	0.002	0.306	0.002	0.442**
	P	0.073	0.001	0.002	0.000	-0.002	0.038	-0.019	0.002	0.231	0.000	0.326**
PH	G	0.002	-0.013	0.010	-0.003	-0.001	0.039	0.015	-0.016	0.000	-0.002	0.031
	P	0.005	0.014	-0.009	-0.002	-0.001	0.009	0.005	-0.004	-0.001	0.000	0.017
SBPP	G	-0.002	-0.004	0.036	-0.023	-0.002	-0.023	0.000	-0.011	-0.008	0.012	-0.025
	P	-0.003	0.003	-0.043	-0.008	-0.001	-0.001	0.001	-0.002	0.011	0.002	-0.043
DFF	G	0.000	0.000	0.004	-0.228	0.002	0.028	0.003	0.020	0.103	0.047	-0.022
	P	0.000	0.000	-0.003	-0.116	0.001	0.006	0.000	0.005	0.077	0.005	-0.024
DFBS	G	0.003	0.000	0.002	0.013	-0.034	0.014	-0.006	0.003	0.094	-0.020	0.068
	P	0.005	0.000	-0.002	0.004	-0.028	0.002	-0.001	0.001	0.072	-0.003	0.051
BPP	G	0.025	-0.002	-0.004	-0.030	-0.002	0.216	-0.050	-0.012	0.317	0.011	0.469**
	P	0.045	0.002	0.001	-0.011	-0.001	0.063	-0.017	-0.002	0.215	0.001	0.297**
SBW	G	-0.022	-0.002	0.000	-0.007	0.002	-0.125	0.087	0.007	-0.340	-0.007	-0.406**
	P	-0.039	0.002	-0.001	-0.002	0.001	-0.029	0.036	0.001	-0.245	-0.001	-0.278**
SPB	G	0.000	-0.002	0.003	0.037	0.001	0.022	-0.005	-0.124	0.058	-0.009	-0.019
	P	-0.002	0.001	-0.002	0.012	0.000	0.003	-0.001	-0.051	0.038	-0.001	-0.003
LFYPP	G	0.013	0.000	0.000	-0.029	-0.004	0.086	-0.037	-0.009	0.797	0.005	0.820**
	P	0.022	0.000	-0.001	-0.012	-0.003	0.018	-0.011	-0.003	0.765	0.001	0.776**
EI	G	-0.001	0.000	-0.005	0.129	-0.008	-0.028	0.007	-0.013	-0.046	-0.083	-0.048
	P	0.000	0.000	0.006	0.042	-0.005	-0.003	0.001	-0.004	-0.029	-0.015	-0.007
Residual effect at genotypic level = 0.258												
Residual effect at phenotypic level = 0.370												

The diagonal values (bold) indicated direct effects and the off diagonal values indicated indirect effects of individual traits to seed cotton yield.

Here, PA= phenotypic acceptability, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, DFBS= days to first boll splitting, BPP= bolls per plant, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness index (%), G= genotypic coefficient of variation, P= phenotypic coefficient of variation and ** indicated significant at 1 % level of probability.

Sympodial branches per plant forecasted positive indirect effect on seed cotton yield per plant via lint fiber yield per plant (0.011) which was in conflict with Rauf *et al.*, 2004; Mawblei *et al.*, 2022. Seeds per boll exhibited positive indirect effect on seed cotton yield per plant via lint fiber yield per plant (0.038), days to 50% flowering (0.012). Patel *et al.*, (2023) reported that seed index imposed a positive and indirect effect on seed cotton yield via lint yield per plant and uniformity index.

The trait earliness index exerted positive indirect effect on seed cotton yield per plant via days to 50% flowering (0.129) but negative indirect effect via lint fiber yield per plant (-0.046), bolls per plant (-0.028) and seeds per boll (-0.013) at genotypic level. Earliness index showed similar directions of positive indirect effect on seed cotton yield per plant via days to 50% flowering (0.042) but negative indirect effect through lint fiber yield per plant (-0.029) at phenotypic level (**Table 11**). Therefore, earliness index phenomenon built up negative correlation with seed cotton yield per plant due other attributing factors at both levels. These findings underscored the intricate interaction of characteristics and their influence on seed cotton yield, offering significant insights for breeding programs focused on improving cotton yield.

In the present study, the element of residual effect in path analysis in yield contributing traits was 0.258 and 0.370 at genotypic level and phenotypic level, respectively. The reduced residual effect suggested that the selected characters for path analysis were suitable and fit in measuring loads of major contributing traits on seed cotton yield. Kadam *et al.*, (2024) observed residual effect of 0.500 at genotypic level and 0.340 at phenotypic level in seed cotton yield per plant contributed by eleven characters of 55 elite cotton genotypes through path analysis. Reddy *et al.*, (2015b) estimated residual effect of path analysis in yield and fiber quality traits was 0.045 at genotypic level and 0.313 at phenotypic level.

4.1.6 Hierarchical cluster analysis based on morphological traits

Hierarchical clustering is a commonly used technique for evaluating genetic diversity (Kakar *et al.*, 2021). This technique initially groups the most similar genotypes, which are then combined based on shared traits. This hierarchical grouping strategy aims to minimize within-group variance and is referred to as Ward's minimal variance method (Ward, 1963). The Euclidean distance coefficients were computed for every 100 upland cotton genotypes on eleven traits. Multivariate analysis revealed that the UPGMA

(unweighted pair group method with arithmetic mean) dendrogram classified the 100 cotton genotypes into eight distinct clusters, namely cluster I, II, III, IV, V, VI, VII, and VIII, (**Figure 7 & Table 12**). In cotton, the cluster group ranged from 2-11 based on hierarchical clustering method (Vishnuvardhan *et al.*, 2023; Abdel-Monaem *et al.*, 2022; Nadeem *et al.*, 2022; Sarwar *et al.*, 2021b, Chapepa *et al.*, 2020; Ullah *et al.*, 2020; Akter *et al.*, 2019; Naik *et al.*, 2016; Pujer *et al.*, 2015).

In the present study, cluster III comprised of the highest number of genotypes (31), followed by cluster VIII (20), and cluster IV (14) (**Table 12**). Genetic divergence among 50 cotton genotypes was also investigated (Naik *et al.*, 2016), categorizing them into 8 clusters where cluster II encompassed the greatest quantity of genotypes (22), succeeded by cluster III (12), cluster I (8), and cluster VII (4), whilst clusters IV, V, VI, and VIII each consisted of a solitary genotype.

Table 13 represents the Cluster means of 11 morphological characteristics in 100 upland cotton genotypes. The current study revealed the average performance of 11 traits across 8 clusters; cluster VI had the greatest means for phenotypic acceptability, bolls per plant, seed cotton yield per plant, and lint fiber yield. Consequently, cluster VI is the optimal selection owing to its elevated mean values. Similar findings were also reported (Jarwar *et al.*, 2019b, Shakeel *et al.*, 2018, Farooq *et al.*, 2017), indicating significant values for yield-contributing and fiber characteristics within the clusters. The genotypes BC-0042, BC-0045, BC-0049 included in Cluster I that exhibited lower mean values for most of the characters but had higher mean values for days to first boll splitting of 133.78 days and single boll weight of 6.01 g. The genotypes RA-2, BC-0024, and BC-0027 were included in Cluster II showed the greatest mean values for the earliness index (92.10%) and the lowest mean values for days to 50% flowering (44.56 days) and single boll weight (4.11 g), respectively. The genotypes in Cluster III and Cluster IV exhibited modest mean values for all traits. The genotypes of cluster V demonstrated the highest mean for seeds per boll (33.98) and the lowest mean for days to first boll splitting (127.50 days). The genotypes RA-5, JA-11/L, BC-0033, BC-0036, BC-0037, BC-0038, BC-0052, and BC-0055 were categorized in cluster VI, exhibited the highest values for various traits, including phenotypic acceptability (8.15), bolls per plant (30.23), seed cotton yield per plant (124.59 g), and lint fiber yield (116.58 g), while demonstrating the lowest values for sympodial

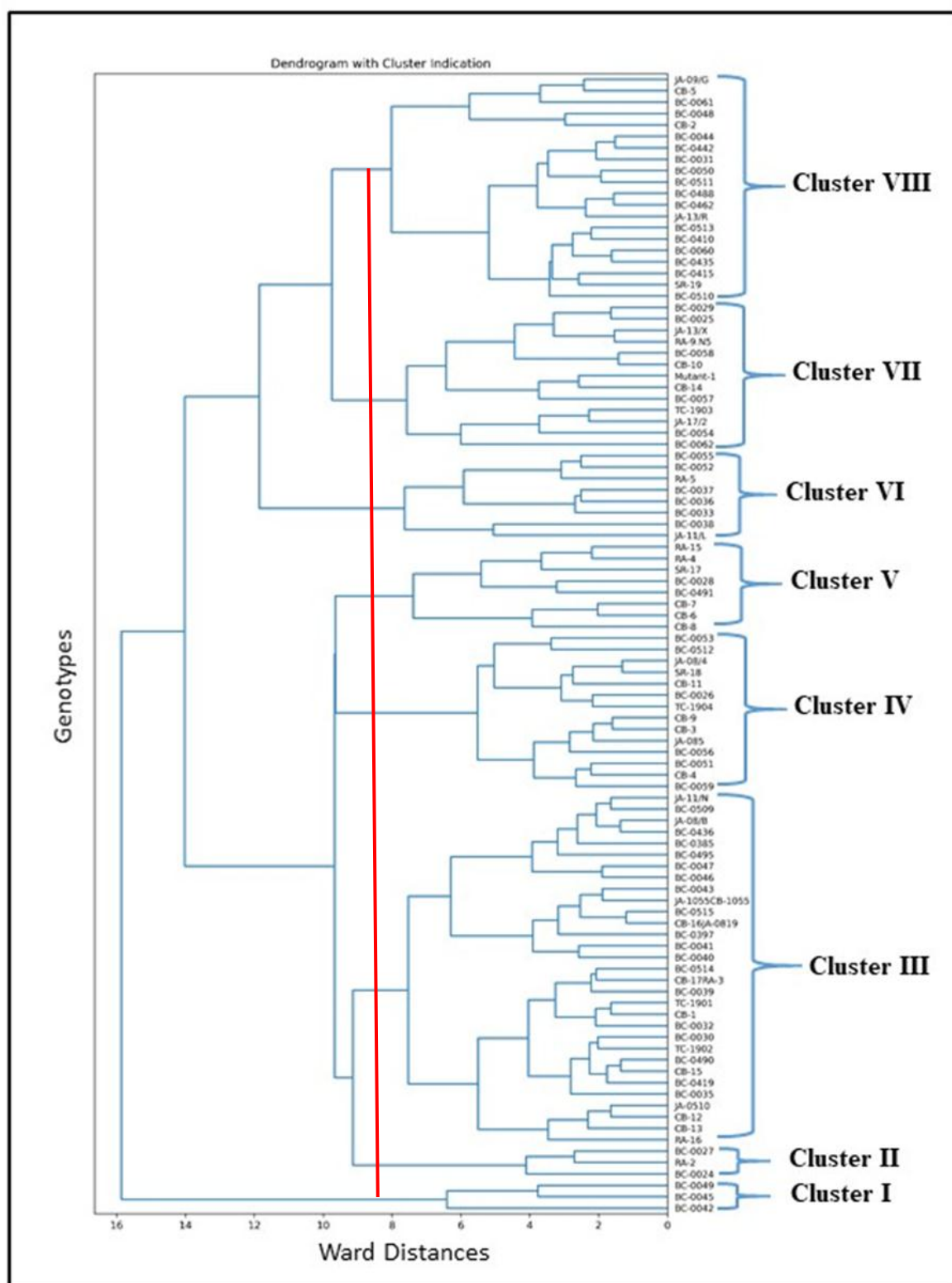


Figure 7. Hierarchical clustering of 100 upland cotton genotypes based on 11 morphological traits

Table 12. Distribution of 100 upland cotton genotypes based on hierarchical clustering method

Cluster group	Number of genotypes per cluster	Distribution of genotypes	Genotypes included
I	3	80, 83, 87	BC-0042, BC-0045, BC-0049
II	3	1, 63, 66	RA-2, BC-0024, BC-0027
III	31	6, 10, 21, 22, 24, 25, 26, 27, 28, 29, 32, 34, 38, 40, 41, 46, 47, 48, 50, 57, 59, 60, 69, 71, 73, 77, 78, 79, 81, 84, 85	RA-16, CB-1, CB-12, CB-13, CB-15, CB-16 JA-0819, CB-17 RA-3, JA- 1055 CB-1055, BC-0385, BC-0397, BC-0419, BC-0436, BC-0490, BC- 0495, BC-0509, BC-0514, BC-0515, JA-11/N, JA-08/B, JA-0510, TC- 1901, TC-1902, BC-0030, BC-0032, BC-0035, BC-0039, BC-0040, BC- 0041, BC-0043, BC-0046, BC-0047
IV	14	8, 12, 13, 18, 20, 44, 55, 56, 62, 65, 89, 91, 94, 97	SR-18, CB-3, CB-4, CB-9, CB-11, BC-0512, JA-08/4, JA-085, TC-190, 4BC-0026, BC-0051, BC-0053, BC-0056, BC-0059
V	8	2, 5, 7, 15, 16, 17, 39, 67	RA-4, RA-15, SR-17, CB-6, CB-7, CB-8, BC-0491, BC-0028
VI	8	3, 51, 72, 74, 75, 76, 90, 93	RA-5, JA-11/L, BC-0033, BC-0036, BC-0037, BC-0038, BC-0052, BC- 0055
VII	13	4, 19, 23, 52, 53, 58, 61, 64, 68, 92, 95, 96, 100	RA-9.N5, CB-10, CB-14, JA-17/2, JA-13/X, Mutant-1, TC-1903, BC-0025, BC-0029, BC-0054, BC-0057, BC-0058, BC-0062
VIII	20	9, 11, 14, 30, 31, 33, 35, 36, 37, 42, 43, 45, 49, 54, 70, 82, 86, 88, 98, 99	SR-19, CB-2, CB-5, BC-0410, BC-0415, BC-0435, BC-0442, BC-0462, BC-0488, BC-0510, BC-0511, BC-0513, JA-13/R, JA-09/G, BC-0031, BC- 0044, BC-0048, BC-0050, BC-0060, BC-0061

branches per plant (17.11) and seeds per boll (28.58). The genotypes of cluster VII revealed the highest mean values for plant height (155.18 cm) and sympodial branches per plant (20.84) respectively. The character days to 50% flowering showed the highest mean values (50.37 days) and lowest mean values for plant height (118.67 cm) and earliness index of 83.40 % under cluster VIII. Cluster I had elevated mean values for days to first boll splitting and single boll weight but reduced mean values for the majority of the traits (**Table 13**).

Based on the objectives, genotypes from cluster II were imperative due to early flowering and higher earliness index. Acceptable values for fiber fineness in cluster I, commendable values for boll weight in cluster III, and favorable values for earliness and staple length in cluster IV were also reported (Saeed *et al.*, 2014).

The **Table 14** displayed both intra-cluster and inter-cluster distances. The highlighted values along the diagonal indicated intra-cluster distances, showing the variability within each cluster. The off-diagonal data illustrated the inter-cluster distances, signifying the genetic divergence between cluster pairs. The maximum inter-cluster distance was noted between clusters I and VI (9.43). The distance between clusters I and VII was 8.53, followed by clusters I and II (8.26). Based on these intra-cluster distances, it was observed that cluster I (2.98) demonstrated the maximum divergence followed by cluster VI (2.93) and cluster V (2.80).

The present study suggested that hybridization involving genotypes from clusters with greater inter-cluster distances, specifically clusters I & VI and I & VII, may be advantageous to produce genetically diverged segregants (**Table 14**). On the other hand, the early maturing genotypes in cluster II such as RA-2, BC-0024 and BC-0049 exhibited moderate inter cluster distance (**Table 12 & Table 14**). Therefore, hybridization between cluster II and VI may produce early maturity in the high yielding varieties with desirable features. A total of 68 genotypes were categorized into 8 clusters based on inter-cluster distances where cluster I and VI exhibited the most significant divergence, followed by cluster V and VI, cluster I and VIII, cluster II and VI (Pujer *et al.*, 2015). According to intra-cluster distances, cluster VI (2.93) exhibited the greatest divergence in our study. Similar findings were also reported (Ali *et al.*, 2019).

Table 13. Cluster means of 11 morphological characteristics in 100 upland cotton genotypes

Cluster	Characters										
	PA	PH (cm)	SBPP	DFE	DFBS	BPP	SBW (g)	SPB	SCYPP (g)	LFYPP (g)	EI (%)
I	3.78 (L)	126.56	18.67	48.00	133.78 (H)	17.67 (L)	6.01 (H)	28.67	115.39 (L)	106.87 (L)	86.45
II	6.73	150.25	19.33	44.56 (L)	133.33	28.56	4.11 (L)	33.33	121.38	113.68	92.10 (H)
III	5.71	118.72	19.30	47.35	129.72	24.46	4.56	31.60	122.35	113.95	85.67
IV	5.41	150.32	19.41	49.17	132.53	24.58	4.93	29.98	122.88	114.79	87.44
V	6.00	150.19	18.51	48.69	127.50 (L)	25.85	5.50	34.00 (H)	122.09	113.24	85.60
VI	8.15 (H)	132.71	17.11 (L)	47.63	132.25	30.23 (H)	4.28	28.58 (L)	124.59 (H)	116.58 (H)	86.55
VII	7.24	155.19 (H)	20.84 (H)	49.39	129.72	27.87	4.42	30.14	123.41	115.35	84.30
VIII	6.17	118.67 (L)	18.88	50.37 (H)	130.10	25.75	4.61	29.62	122.89	115.16	83.40 (L)

Here, PA= phenotypic acceptability, PH= plant height (cm), SBPP= sympodial branches per plant, DFE= days to 50% flowering, DFBS= days to first boll splitting, BPP= bolls per plant, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness index, H= highest and L= lowest.

Table 14. Intra (diagonal) and inter (off-diagonal) cluster distances in 100 upland cotton genotypes

Cluster	I	II	III	IV	V	VI	VII	VIII
I	2.98	8.26	6.94	6.85	7.30	9.43	8.53	7.57
II		1.99	4.85	5.02	5.77	5.58	5.54	6.00
III			2.13	3.73	4.42	5.34	4.45	3.77
IV				2.10	4.62	5.30	4.36	3.97
V					2.80	6.26	5.30	4.94
VI						2.93	5.42	5.05
VII							2.74	4.39
VIII								2.36

4.1.7 Principal Component Analysis among the 100 upland cotton genotypes

Principal component analysis (PCA) was performed to allocate the total variance into principal components, facilitating the identification of superior germplasm based on genetic diversity for breeding programs (Shakeel *et al.*, 2015, Nazir *et al.*, 2013). In PCA, the eigen value represents the component that elucidates variance for a particular character. It is deemed more appropriate technique as it reveals the greatest contributor significance to the total variation along each differentiation axis according to Sharma, 2006. Numerous researchers have employed this method to quantify the diversity within germplasm (Nadeem *et al.*, 2022). Principal Component Analysis (PCA) was conducted using mean values of 100 upland cotton genotypes on eleven characters. Here, only first five principal components (PCs) had eigen values greater than 1.00 with maximum cumulative variance of 73.087% (**Table 15**).

In the context of PCA, a higher eigen value associated with a component signified that it captured more variance in the data, making it more representative of the underlying structure of the data set (Ullah *et al.*, 2020). In the present study, the variance and eigen values diminished markedly from PC1 (27.095%, 2.980) to PC5 (9.311%, 1.024). This means that after the fifth principal component (PC5), the additional principal components contribute very little additional variation to the data set. Consequently, only the initial five principal components were taken into account. The current results aligned with the findings where PCA resulted in only four components, with eigen values over 1, accounting for 65% of the entire variability in 102 elite cotton genotypes (Ullah *et al.*, 2020; Kumari & Gunasekaran, 2019; Shah *et al.*, 2018; Saeed *et al.*, 2014). On the other hand, only PC1 and PC2 significantly contributed to their correlation analysis, as the remaining principal components account for minimal data information also reported (Chunthaburee *et al.*, 2016). In first principal component (PC1), the phenotypic acceptability (0.799), bolls per plant (0.786), single boll weight (-0.742), seed cotton yield per plant (0.756) and lint fiber yield per plant (0.748) bears higher co-efficient values greater than 0.3. The coefficients of the proper vector exceeding 0.3, irrespective of direction positive or negative indicate significant effects deemed important, whereas traits below 0.3 lack substantial impact on overall variation (Raji, 2003). The PC1 explained 27.095% of the total variability with eigen value 2.98. Significant contribution of the first main component to overall variability in the analysis of several traits was also reported (Ali *et al.*, 2019).

Table 15. Arrangement of principal components against 11 morphological characteristics in 100 upland cotton genotypes

Characters	PC1	PC2	PC3	PC4	PC5
Phenotypic acceptability	0.799	0.065	-0.011	-0.354	0.233
Plant height (cm)	0.042	-0.055	0.789	-0.117	0.082
Sympodial branches per plant	-0.047	-0.275	0.662	0.136	0.095
Days to 50% flowering	0.089	-0.796	-0.025	0.115	0.143
Days to first boll splitting	0.119	0.334	0.204	0.401	0.636
Bolls per plant	0.786	-0.048	0.099	-0.373	0.145
Single boll weight (g)	-0.742	-0.056	0.172	0.272	-0.078
Seeds per boll	0.044	0.245	0.424	-0.253	-0.588
Seed cotton yield per plant (g)	0.756	0.061	0.001	0.471	-0.268
Lint fiber yield per plant (g)	0.748	-0.011	0.027	0.523	-0.277
Earliness index (%)	-0.100	0.839	0.057	0.057	0.091
Eigen value	2.980	1.602	1.326	1.107	1.024
Percentage of variance (%)	27.095	14.563	12.054	10.064	9.311
Cumulative percentage (%)	27.095	41.658	53.712	63.776	73.087

Here, PC= principal component.

The second main component (PC2) explained 14.563% of the overall variability with 1.602 eigen value. Here, the traits days to 50% flowering (-0.796), days to first boll splitting (0.334) and earliness index (0.839) had higher co-efficient values. The third component (PC3) accounted 12.054% to the variance, in which the traits plant height (0.789), sympodial branches per plant (0.662) and seeds per boll (0.424) contributed mostly.

The fourth (PC4) and fifth principal components (PC5) accounted for approximately 10.064% and 9.311% of the total variability, with eigen values of 1.107 and 1.024, respectively. In PC4 the character's phenotypic acceptability (-0.354), days to first boll splitting (0.401), bolls per plant (-0.373), seed cotton yield per plant (0.471) and lint fiber yield per plant (0.523) contributed mostly whereas in PC5, the variable days to first boll splitting (0.636) and seeds per boll (-0.588) played vital role.

Isong *et al.*, 2017 reported that the first PC explained the highest variability of 22.54% and was most related to seed cotton yield, boll weight, number of bolls per plant, uniformity ratio, number of sympodia and plant height, while the second and third PCs had variability of 13.1% and 10.7% respectively and were more associated with days to flowering, days to first bursting, 2.5% span length and seed index, whatsoever the rest PCs were more associated with, days to 50% flowering, number of seeds per boll and bundle strength.

4.1.8 Biplot analysis among the 100 upland cotton genotypes

A biplot illustrated the interaction between genotype and traits, as well as the correlation among traits, utilizing the mean values of cotton genotypes. The genotypes near the origin exhibited greater similarity with each other, while those further from the origin displayed maximum variation and reduced similarity (Bhatti *et al.*, 2020a,b). Again, the characters exhibited a positive correlation if the angle between their vectors is less than 90 degree, and a negative correlation if the angle exceeds 90 degree whereas the trait vectors that are almost orthogonal exhibit independence (Zafar *et al.*, 2022).

Positive loadings were seen in PC1 and PC2 for the traits phenotypic acceptability, days to first boll splitting, seeds per boll and seed cotton yield per plant (g) (**Figure 8**). The traits phenotypic acceptability and seed cotton yield per plant were strongly correlated and associated with the genotypes JA-11/L (51), RA-5 (3), BC-0033 (72), BC-0036 (74),

BC-0037 (75) among others. The genotype CB-12 (21), CB-17, RA-3 (26), BC-0056 (94) and SR-17 (7) had a strong correlation with seeds per boll and the days to first boll splitting.

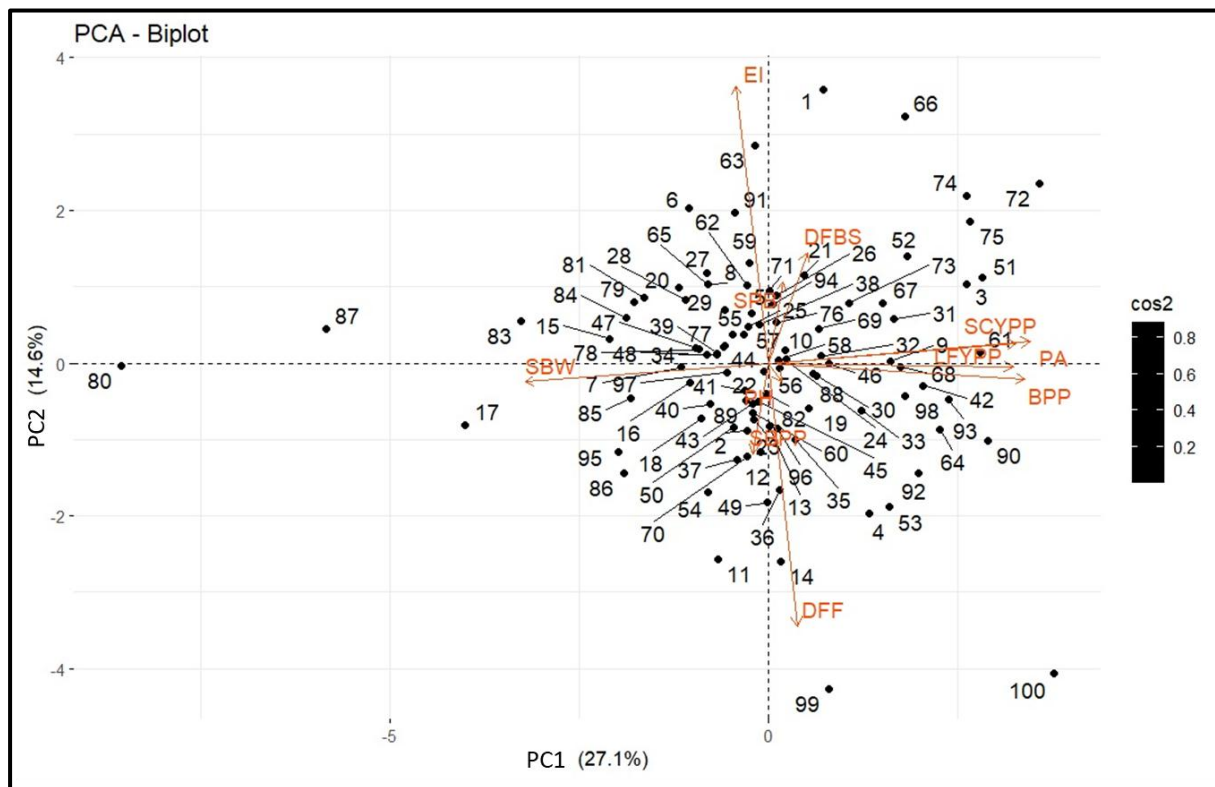


Figure 8. PCA biplot of 11 yield and yield contributing traits in 100 upland cotton genotypes

Here, PA= phenotypic acceptability, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, DFBS= days to first boll splitting, BPP= bolls per plant, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness index (%), PC= principal component and the number showing genotypes.

Traits such as plant height, days to 50% flowering, bolls per plant and lint fibre yield per plant were associated with positive PC1 and negative PC2 scores. They exhibited a robust link with each other. In this instance, the genotypes CB-10 (19), BC-0029 (68), BC-0510 (42), BC-0052 (90) and BC-0055 (93) showed a preference for these traits. Additionally, PC2 exhibited positive loading, but PC1 demonstrated negative score for the trait earliness index.

The genotypes BC-0024 (63), BC-0027 (66), RA-2 (1) and BC-0053 (91) were strongly associated with the earliness trait performances. The parameters of sympodial branches per plant and single boll weight (g) were located on the negative axis of both PC1 and PC2. The features were interrelated and associated with the genotype numbers JA-09G (54), BC-0488(37), BC-0031 (70), CB-3 (12), BC-0042 (80), BC-0049 (87), BC-0045 (83) and CB-8 (17) among others. The unique scatter behavior of the genotypes GP-1, GP-39, GP-21, GP-8, GP-38, and VH-259 (check) located at the vertex of the polygon in the biplot analysis of 102 upland cotton genotypes was demonstrated (Jamil *et al.*, 2022). A comparable correlation among plant height, ginning, lint index, seed index, and cotton seed weight was also reported by Shaheen *et al.*, 2021. Again, high amount of variability among most of the studied yield and yield contributing traits except root traits in PCA-biplot was estimated by Rathinavel K., (2018). Overall, a high range of variability existed in 100 cotton genotypes prevailing greater divergence and may be useful for an effective cotton breeding programs.

4.2 Experiment II: Evaluation of 45 upland cotton genotypes for revealing early performances and measuring genetic diversity

The primary objective of the investigation was to select early boll harvesting with high seed cotton yield contributing traits and genetically diverged genotypes to develop desirable early varieties through judicious and effective breeding programs. Therefore, genetic parameters and character associations were studied and then 100 genotypes were grouped in eight clusters by hierarchical and PCA based on seed cotton yield and its ten related characters. From where 45 early and high seed cotton yielding genotypes were selected (**Table 3**) and evaluated for early performances.

4.2.1 Analysis of variance (ANOVA) of the 45 upland cotton genotypes

The analysis of variance showed highly significant differences among the 45 upland cotton genotypes across all ten morphological traits studied, as summarized by the mean sum of squares in **Table 16**. The strong significance ($p < 0.001$) of genotype effects on plant height, sympodial branches per plant, days to 50% flowering, number of bolls per plant, boll bursting percentage, single boll weight, seeds per boll, seed cotton yield per plant, lint fiber yield per plant and earliness index (%) highlighted the considerable genetic variability present in the material tested. These results suggested that these traits were under predominantly genetic control, increasing the likelihood that high-yielding genotypes could be identified and selected for cotton breeding. These traits are key for developing compact, early-maturing cotton varieties, aligning with previous research emphasizing their importance for high yield and adaptation (Hu *et al.*, 2022; Soomro *et al.*, 2015; Ahmed *et al.*, 2008).

The coefficient of variation (CV%) values for all traits were generally low, ranging from a minimal value for the earliness index to 9.31% for sympodial branches per plant, indicated good precision was present in genotypes randomizations in three blocks and unbiased samplings for data collection. In addition, these low CV values indicated that the experimental data were precise and reliable, with minimal influence from environmental variations.

The grand mean of the yield components established a baseline for the overall performance evaluation of the genotypes. These observations were consistent with previous research reporting significant genetic variation for yield-related traits in cotton

Table 16. Mean squares (MS) derived on 10 morphological characters in 45 upland cotton genotypes

Traits	Mean Sum of Square					
	Genotype	Replication	Error	CV%	Mean Range	Grand Mean $\pm$ SE
Plant height (cm)	582.67***	2.56	2.75	1.25	109.60-177.60	133.15 $\pm$ 0.96
Sympodial branches per plant	20.76***	2.76	3.13	9.31	13.00-26.00	19.00 $\pm$ 1.02
Days to 50% flowering	42.67***	0.36	2.09	2.47	49.00-64.00	58.61 $\pm$ 0.83
Bolls per plant	24.34***	7.18	2.53	6.32	20.00-33.40	25.18 $\pm$ 0.92
Boll bursting percentage (%)	526.69***	6.71	4.04	2.52	51.12-98.00	79.81 $\pm$ 1.16
Single boll weight (g)	0.48***	0.13	0.10	6.64	4.00-5.89	4.75 $\pm$ 0.18
Seeds per boll	37.50***	2.61	2.29	5.86	19.00-35.65	25.78 $\pm$ 0.87
Seed cotton yield per plant (g)	202.48***	0.90	3.12	1.54	100.15-129.93	114.88 $\pm$ 1.02
Lint fiber yield per plant (g)	197.79***	0.83	3.39	1.82	86.66-115.00	101.17 $\pm$ 1.06
Earliness index (%)	54.36***	3.91*	1.0	1.14	75.98-96.00	87.93 $\pm$ 0.58

Here, *** indicated significant at 0.1% levels of probability, CV= co-efficient of variation, SE= standard error of mean, LSD= least significant difference

(Mawblei *et al.*, 2022; Saraswat *et al.*, 2022; Ali, 2019; Alam, 2018; Amin, 2018; Monisha *et al.*, 2018; Eswari *et al.*, 2017). On average, the plants reached a height of 133.15 ± 0.96 cm, and the average number of bolls per plant was 25.18 ± 0.92 . Days to 50% flowering ranged from 49.00-64.00 days whereas. Saraswat *et al.* (2022) and Baloch *et al.* (2014) reported days to first flower expressed changed from 51.6 to 66.6 days and 90 and 120 days, respectively. Therefore, I got more early flowering genotypes than others. The genotypes also demonstrated promising productivity, with a mean seed cotton yield of 114.88 ± 1.02 g per plant and a lint fiber yield of 101.17 ± 1.06 g per plant (**Table 16**). The current study recorded more yield than Vadodaria *et al.* (2024) and Modi *et al.* (2020), who found 80.07 g per plant and 86.4g per plant yield, respectively. Additionally, the high average boll bursting percentage of $79.81 \pm 1.16\%$ and an earliness index of $87.93 \pm 0.58\%$ suggested that these genotypes might be well adapted for cultivation within a short growing period. Meanwhile, Seyyed *et al.* (2022b) obtained 73.5% earliness.

The highly significant mean squares observed for genotypes across all evaluated traits clearly demonstrated the presence of substantial genetic diversity among the selected upland cotton genotypes (Vadodaria *et al.*, 2024; Divya *et al.*, 2023; Subalakhshmi *et al.*, 2023; Vishnuvardhan *et al.*, 2023; Balarabe *et al.*, 2022; Meena *et al.*, 2022b; Nadeem *et al.*, 2022; Bhatti *et al.*, 2020a,b; Mahdi & Emam, 2020; Jarwar *et al.*, 2019b; Khan *et al.*, 2003). This finding was particularly valuable in the context of diversity analysis, as it highlighted the potential for breeding programs to select superior genotypes based on these traits. Genetic variability is essential for any successful breeding effort, as it provides the foundation materials necessary for effective selection and improvement (Baloch *et al.*, 2015).

4.2.2 Mean performance of 45 upland cotton genotypes

The mean values for ten morphological and yield-related traits across 45 upland cotton genotypes were presented in **Table 17**. The Least Significant Difference (LSD) values at the bottom of the table indicated statistically significant differences among genotypes for all traits studied.

Significant variation was observed in seed cotton yield per plant (SCYPP), with genotype BC-0491 producing the highest yield at 127.71 g, significantly out performing all other genotypes. This was followed by BC-0027 (127.56 g), BC-0510 (127.25 g),

BC-0512 (126.98 g), JA-13/X (126.80 g) and RA-5 (126.61 g) while BC-0057 had the lowest yield (101.5 g). These marked differences highlighted the potential for selecting high-yielding genotypes. Lint fiber yield per plant (LFYPP) followed a similar pattern, where genotypes with high seed cotton yield also exhibited superior lint yield. RA-5 led with 113.63 g, followed by BC-0491 at 113.40 g, BC-0510 at 113.06 g, JA-13/X at 112.77 g, BC-0512 at 112.61 g and BC-0027 at 112.33 g, respectively. The lowest LFYPP was recorded in BC-0057 (88.12 g). The strong positive relationship between seed cotton yield and lint fiber yield is essential in breeding for productive cotton varieties.

The number of bolls per plant (BPP) is a key yield determinant. TC-1903 recorded the highest count (31.93), followed by Mutant-1 (30.93), BC-0510 (30.47), JA-17/2 (30.27), BC-0027 (30.13) and BC-0512 (30.0) whereas BC-0059 produced the fewest bolls (22.40). These significant differences aligned with the importance of bolls per plant in end product, yield. Single boll weight (SBW) varied considerably across genotypes, with BC-0027 exhibiting the heaviest bolls (5.81 g) and BC-0052 the lightest (4.23 g).

Significant variation was also found in plant height (PH), ranging from the tallest genotype BC-0058 (176.47 cm) to the shortest BC-0049 (112.2 cm). This variability is important for breeding cotton adapted to specific cropping season, especially where lodging risk from excessive rainfall is a concern.

Sympodial branches per plant (SBPP) followed a similar trend, with TC-1903 (24.33), BC-0510 (24.0), BC-0512 (23.67), JA-13/X (23.67) and BC-0027 (23.0) having the highest numbers. A greater number of sympodial branches generally provides more fruiting sites, supporting higher yield.

Days to 50% flowering (DFF), a critical indicator of maturity, varied significantly with RA-2 (50.0 days), BC-0512 (51.0 days), BC-0491 (51.33 days) flowering earliest whereas BC-0029, BC-0048, BC-0058 and BC-0061 of the latest (60.0 days). This variation supported the selection of genotypes for different maturity durations.

The earliness index (EI) further confirmed these differences, where BC-0510(95.0%), BC-0491 (94.33%), RA-2 (94.08%) and RA-9.N5 (94.0%) registered the highest value signaling strong potential as an early-maturing genotype, while BC-0060 (76.56%) and BC-0061 (77.12%) had the lowest.

Table 17. Mean values based on phenotypic expressions of 10 morphological characteristics in 45 upland cotton genotypes

SL.	Genotype	PH (cm)	SBPP	DFF	BPP	BBP (%)	SBW (g)	SPB	SCYPP (g)	LFYPP (g)	EI (%)
1	RA-2	117.47 yz	19.33 f-l	50.00 k	28.13 c-e	97.16 a	5.26 b-e	30.30 bc	124.95 ab	111.03 a-c	94.08 a-c
2	RA-5	120.00 w-y	20.33 d-j	52.00 h-k	27.13 d-f	95.64 a	5.35 a-d	29.63 b-e	126.61 a	113.63 a	92.67 c-e
3	RA-9.N5	118.90 x-z	19.67 e-k	53.00 g-j	29.13 b-d	96.27 a	5.40 a-c	31.73 ab	123.53 bc	109.45 c-e	94.00 a-c
4	CB-2	134.93 i-k	15.67 n-q	61.67 ab	21.33 o	88.53 de	4.40 f-i	29.60 b-e	120.27 d-f	107.77 def	80.21 m
5	CB-6	124.13 v	14.33 q	57.00 ef	23.00 i-o	92.23 bc	4.84 d-g	23.60 k-p	121.99 cd	109.84 b-d	88.35 f-h
6	CB-7	131.40 n-q	15.33 o-q	59.33 b-e	23.20 i-o	89.70 cd	4.49 f-i	27.20 e-h	117.59 f-i	105.48 f-h	86.35 ij
7	CB-8	134.20 i-m	15.33 o-q	60.33 a-d	25.00 f-k	88.51 de	4.56 f-i	27.73 d-g	120.52 de	107.83 d-f	85.11 jk
8	CB-10	130.93 o-r	19.33 f-l	58.67 de	25.27 f-j	89.67 cd	4.51 f-i	26.33 g-j	117.73 e-h	105.20 f-i	86.75 hi
9	CB-14	133.53 k-o	20.00 e-j	57.00 ef	26.53 e-g	85.55 e-g	4.77 e-h	33.63 a	117.20 g-i	100.57 jk	86.35 ij
10	BC-0491	127.00 u	22.33 a-e	51.33 i-k	28.13 c-e	96.67 a	5.36 a-c	29.67 b-d	127.71 a	113.40 a	94.33 ab
11	BC-0510	127.53 tu	24.00 ab	52.00 h-k	30.47 a-c	96.22 a	5.56 ab	28.14 c-g	127.25 a	113.06 a	95.00 a
12	BC-0512	127.27 u	23.67 a-c	51.00 jk	30.00 a-c	94.00 ab	5.35 a-d	29.97 b-d	126.98 a	112.61 ab	94.67 ab
13	JA-11/L	136.47 ij	16.67 l-q	52.00 h-k	24.27 g-n	87.77 de	4.42 f-i	24.67 i-n	116.27 hi	102.05 j	89.11 f
14	JA-17/2	130.33 p-s	22.00 a-f	53.67 g-i	30.27 a-c	86.60 d-f	5.26 b-e	30.1 b-d	125.91 ab	111.67 a-c	92.33 de
15	JA-13/X	131.73 m-p	23.67 a-c	54.67 fg	30.13 a-c	89.64 cd	5.55 ab	29.43 b-f	126.80 a	112.77 ab	92.00 de
16	Mutant-1	145.53 fg	16.67 l-q	59.00 c-e	30.93 ab	80.32 j-l	4.46 f-i	31.03 b	120.63 d	106.59 ef	88.25 f-h
17	TC-1903	130.13 p-t	24.33 a	54.00 gh	31.93 a	95.38 ab	5.27 b-e	31.03 b	125.78 ab	112.12 a-c	91.33 e
18	TC-1904	141.20 h	15.33 o-q	61.33 a-c	21.87 no	82.23 h-j	4.65 f-i	24.33 j-o	108.12 k-m	94.61 lm	88.25 f-h
19	BC-0024	144.13 g	18.00 i-o	61.00 a-d	22.00 m-o	76.21 m-o	4.51 f-i	22.00 o-r	107.56 k-n	93.87 lm	89.67 f
20	BC-0025	154.27 d	19.00 g-m	62.00 a	24.13 g-n	78.58 k-n	4.45 f-i	27.00 f-i	117.54 f-i	103.14 g-j	86.11 ij
21	BC-0026	136.73 i	18.00 i-o	59.33 b-e	23.27 i-o	67.44 q	4.78 e-h	26.00 g-k	103.16 o-q	90.33 n-q	89.12 f
22	BC-0027	122.13 vw	23.00 a-d	53.33 g-j	30.13 a-c	94.00 ab	5.81 a	31.50 ab	127.56 a	112.33 a-c	93.25 b-d
23	BC-0028	126.93 u	19.00 g-m	61.00 a-d	24.40 g-n	75.52 n-p	4.66 f-i	25.00 h-m	115.91 hi	102.62 h-j	89.59 f
24	BC-0029	147.87 ef	19.33 f-l	62.00 a	23.33 i-o	86.34 e-g	4.42 f-i	24.29 j-o	107.74 k-n	93.77 lm	88.30 f-h
25	BC-0033	120.6 wx	20.00 e-j	60.33 a-d	24.27 g-n	65.36 qr	4.69 f-i	24.33 j-o	105.17 n-p	90.74 m-p	88.63 fg

Table 17. Mean values based on phenotypic expressions of 10 morphological characteristics in 45 upland cotton genotypes

SL.	Genotype	PH (cm)	SBPP	DFF	BPP	BBP (%)	SBW (g)	SPB	SCYPP (g)	LFYPP (g)	EI (%)
26	BC-0036	123.73 v	20.33 d-j	61.33 a-c	24.87 f-l	88.05 de	4.68 f-i	22.00 o-r	108.23 j-l	94.04 lm	89.35 f
27	BC-0037	116.33 z	21.33 b-g	60.00 a-d	24.53 g-m	84.09 f-i	4.42 f-i	21.40 p-r	108.36 j-l	94.13 lm	89.45 f
28	BC-0038	117.47 yz	20.00 e-j	61.33 a-c	24.27 g-n	86.03 e-g	4.28 hi	24.67 i-n	111.08 j	98.35 k	89.55 f
29	BC-0042	110.33 a	18.67 g-m	60.67 a-d	24.4 g-n	61.47 s	4.59 f-i	25.20 h-l	119.45 d-g	105.93 fg	88.12 f-h
30	BC-0045	134.00 j-n	18.00 i-o	61.00 a-d	23.53 h-o	78.89 k-m	4.46 f-i	25.00 h-m	117.09 g-i	102.51 h-j	88.23 f-h
31	BC-0046	132.13 l-p	18.33 h-n	61.33 a-c	23.00 i-o	72.59 p	4.61 f-i	22.33 n-r	107.62 k-n	93.67 l-n	89.23 f
32	BC-0047	128.93 q-u	20.67 d-i	60.33 a-d	22.47 k-o	85.30 e-h	4.62 f-i	23.00 l-p	115.09 hi	101.10 jk	87.12 g-i
33	BC-0048	127.93 s-u	15.00 pq	62.00 a	23.07 i-o	81.68 i-k	4.52 f-i	25.87 ^{g-k}	114.87 i	102.38 ij	83.25 l
34	BC-0049	112.20 a	15.00 pq	60.00 a-d	23.73 h-o	66.38 qr	4.60 f-i	24.00 j-o	102.68 pq	89.33 o-q	88.25f-h
35	BC-0050	135.60 i-k	17.00 k-q	61.00 a-d	24.07 g-n	77.46 l-n	4.36 g-i	23.70 k-p	106.88 k-n	94.31 lm	86.23 ij
36	BC-0052	133.67 k-n	15.67 n-q	60.67 a-d	23.20 i-o	83.13 g-j	4.23 i	27.00 f-i	108.02 k-n	93.82 lm	84.25 kl
37	BC-0053	148.27 e	16.33 m-q	60.33 a-d	23.67 h-o	55.56 t	4.93 c-f	23.70 k-p	103.42 o-q	90.33 o-q	85.00 jk
38	BC-0054	136.47 ij	19.00 g-m	61.33 a-c	23.00 i-o	61.23 s	4.73 f-i	23.00 l-p	105.87 l-o	92.49 l-n	84.12 kl
39	BC-0055	134.67 i-l	17.67 j-p	60.00 a-d	25.33 f-i	78.59 k-n	4.64 f-i	21.33 p-r	109.24 jk	94.99 l	86.23 ij
40	BC-0057	158.87 c	17.00 k-q	61.33 a-c	22.73 j-p	53.38 t	4.84 d-g	22.00 o-r	101.50 q	88.12 pq	88.12 f-h
41	BC-0058	176.47 a	20.33 d-j	62.00 a	23.93 h-n	55.07 t	4.26 i	20.40 qr	107.41 k-n	93.71 lm	86.25 ij
42	BC-0059	145.53 fg	19.33 f-l	61.33 a-c	22.40 l-o	73.30 op	4.70 f-i	20.00 r	105.31 m-p	91.89 l-p	87.23 g-i
43	BC-0060	128.33 r-u	19.67 e-k	61.00 a-d	23.33 i-o	63.87 rs	4.68 f-i	22.67 m-q	115.36 hi	101.39 j	76.56 n
44	BC-0061	121.80 vw	21.00 c-h	62.00 a	23.33 i-o	55.58 t	4.36 g-i	21.33 p-r	105.95 l-o	92.12 l-o	77.12 n
45	BC-0062	173.77 b	20.33 d-j	61.67 ab	26.00 e-h	54.38 t	4.51 f-i	23.00 l-p	115.58 hi	101.58 j	79.10 m
SD		13.90	2.99	3.93	3.12	13.26	0.47	3.72	8.28	8.20	4.31
LSD (5%)		2.69	2.87	2.35	2.58	3.26	0.51	2.45	2.87	2.99	1.62

Here, the mean values bearing same letter (s) didn't differ significantly at $p < 0.05$, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, BPP= bolls per plant, BBP= boll bursting percentage (%), SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness Index (%), SD= standard deviation and LSD (5%)= least significance difference at 5% level of probability.

The significant variability detected among the 45 upland cotton genotypes across all studied traits aligned with previous variance analyses (**Table 16**) and confirmed a major genetic base for practical breeding. This variability allowed breeders to select superior genotypes, particularly for yield-related traits and their components.

A clear positive relationship emerged between seed cotton yield and yield components such as bolls per plant and single boll weight. Top-yielding genotypes like BC-0027, BC-0510, JA-13/X and BC-0512 also excelled in these traits (**Table 17**). This was consistent with established studies emphasizing the importance of bolls per plant and boll weight as the primary contributors to yield (Alehegn *et al.*, 2020; Khan *et al.*, 2015). Accordingly, breeding programs should prioritize these traits.

The results also emphasized the typical trade-off between yield and earliness. High-yielding genotypes such as BC-0491, BC-0510, BC-0512 tended to early flowering and had higher earliness indices. Early-maturing genotypes are vital in regions prone to late-season stresses like drought and insect pressure (Kiani *et al.*, 2007). The genotype BC-0510, with the highest earliness index, represented a valuable resource for breeding cotton varieties adapted to short-season environments. Substantial variation in plant height and sympodial branching offers opportunities for breeding cotton ideotypes. Managing plant height to prevent lodging while maintaining yield is a key breeding objective. Genotypes such as BC-0027, BC-0512, BC-0491 and BC-0510 combined shorter stature and early flowering, could serve as promising parents in breeding programs aimed at developing compact, early, and high-yielding varieties.

4.2.3 Dunnett's test for comparing the performance of 45 upland cotton genotypes against two standard checks

The results of a Dunnett's test comparing the mean performance of morphological traits in 45 upland cotton genotypes against two standard check varieties, CB-14 and Mutant-1 were displayed in **Table 18**. The analysis provided a clear indication of which genotypes showed significant differences in key morphological traits.

In plant height (PH), several genotypes exhibited significantly different plant heights compared to the checks. Genotypes such as BC-0042, BC-0049, BC-0037, BC-0038 and RA-2 were all significantly shorter than both checks, with differences reaching up to -35.20^{***} cm compared to Mutant-1. Conversely, genotypes like BC-0058 and BC-

0062 were significantly taller, with the latter showing a highly significant increase of 30.93*** cm compared to Mutant-1.

In sympodial branches per plant (SBPP), the number of sympodial branches varied significantly among genotypes. Genotype TC-1903, BC-0510, BC-0512 and JA-13/X showed a highly significant increase of 7.67***, 7.33*** and 7.0*** compared to both checks, suggesting a higher potential for boll production.

In days to 50% flowering (DFF), many genotypes flowered significantly earlier than the checks. The most notable differences were seen in genotypes RA-2, BC-0512, BC-0491, BC-0510 and JA-11/L which showed highly significant reductions in days to flowering, with values of -9.0***, -8.0***, -7.67*** and -7.0*** days, respectively. Conversely, genotypes like BC-0458, BC-0061, BC-0029 and BC-0025 were significantly later to flower.

In bolls per plant (BPP), this trait, a key component of yield, showed significant differences. Genotype TC-1903 had significant increases of 5.40** compared to CB-14. Other genotypes, such as BC-0048, showed a highly significant reduction in BPP.

Furthermore, the boll bursting percentage (BBP) also showed significant variation. Genotype RA-2, BC-0491 and RA-9.N5 exhibited a highly significant increase of 16.84***, 16.35*** and 15.95*** compared to both checks. Conversely, many genotypes showed significant negative differences, with some, like genotype BC-0057, BC-0062 and BC-0058, having highly significant reductions of -26.94***, -25.94*** and -25.25***.

The Dunnett's test results in **Table 18** highlighted significant variability among 45 upland cotton genotypes in key morphological and developmental traits when compared to the standard checks CB-14 and Mutant-1. While some genotypes demonstrated favorable traits such as reduced plant height and earlier flowering, which might be advantageous for certain agronomic practices, these differences must be interpreted cautiously in the context of overall crop performance.

For instance, although shorter plants like BC-0042 might be less prone to lodging, excessively reduced height could limit biomass and potentially affect yield components (Singh *et al.*, 2020). Similarly, genotypes showing significantly earlier flowering, such as RA-2, might facilitate early maturity and crop rotation benefits; yet, early flowering

Table 18. Dunnett's test performed on morphological trait means to examine differences between each of the 45 upland cotton genotypes and two checks (CB-14 and Mutant-1)

SL.	Genotype	PH (cm)		SBPP		DFF		BPP		BBP (%)	
		CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1
1.	RA-2	-16.07***	-28.07***	-0.67	2.67	-7.0***	-9.0***	1.60	-2.80	11.61***	16.84***
2.	RA-5	-13.53***	-25.53***	0.33	3.67	-5.0**	-7.0***	0.60	-3.80	10.09***	15.32***
3.	RA-9.N5	-14.63***	-26.63***	-0.33	3.00	-4.0*	-6.0***	2.60	-1.80	10.72***	15.95***
4.	CB-2	1.40	-10.60***	-4.33 .	-1.0	4.67**	2.67	-5.20**	-9.60	2.98	8.21***
5.	CB-6	-9.40***	-21.40***	-5.67**	-2.33	0.00	-2.0	-3.53	-7.93***	6.68**	11.91***
6.	CB-7	-2.13	-14.13***	-4.67*	-1.33	2.33	0.33	-3.33	-7.73***	4.15	9.38***
7.	CB-8	0.67	-11.33***	-4.67*	-1.33	3.33	1.33	-1.53	-5.93***	2.96	8.19***
8.	CB-10	-2.60	-14.60***	-0.67	2.67	1.67	-0.33	-1.27	-5.67**	4.12	9.35***
9.	CB-14	-	-12.00***	-	3.33	-	-2.0	-	-4.40*	-	5.23 .
10.	BC-0491	-6.53***	-18.53***	2.33	5.67**	-5.67***	-7.67***	1.60	-2.80	11.12***	16.35***
11.	BC-0510	-6.00***	-18.00***	4.00	7.33***	-5.0**	-7.0***	3.93	-0.47	10.67***	15.90***
12.	BC-0512	-6.27***	-18.27***	3.67	7.0***	-6.0***	-8.0***	3.47 .	-0.93	8.45***	13.68***
13.	JA-11/L	2.93	-9.07***	-3.33	0.00	-5.0**	-7.0***	-2.27	-6.67***	2.22	7.45***
14.	JA-17/2	-3.20	-15.20***	2.0	5.33*	-3.33	-5.33***	3.73	-0.67	1.05	6.28**
15.	JA-13/X	-1.80	-13.80***	3.67	7.0***	-2.33	-4.33*	3.60	-0.80	4.09	9.32***
16.	Mutant-1	12.00***	-	-3.33	-	2.0	-	4.40*	-	-5.23 .	-
17.	TC-1903	-3.40	-15.40***	4.33 .	7.67***	-3.0	-5.0**	5.40**	1.0	9.83***	15.06***
18.	TC-1904	7.67***	-4.33*	-4.67 *	-1.33	4.33*	2.33	-4.67*	-9.07***	-3.32	1.91
19.	BC-0024	10.60***	-1.40	-2.00	1.33	4.0*	2.0	-4.53*	-8.93***	-9.34***	-4.11
20.	BC-0025	20.73***	8.73***	-1.00	2.33	5.0**	3.0	-2.40	-6.80***	-6.97**	-1.74
21.	BC-0026	3.20	-8.80***	-2.00	1.33	2.33	0.33	-3.27	-7.67***	-18.11***	-12.88***
22.	BC-0027	-11.40***	-23.40***	3.00	6.33**	-3.67 .	-5.67***	3.60	-0.80	8.45***	13.68***
23.	BC-0028	-6.60***	-18.60***	-1.00	2.33	4.0*	2.0	-2.13	-6.53***	-10.03***	-4.80 .
24.	BC-0029	14.33***	2.33	-0.67	2.67	5.0**	3.0	-3.20	-7.60***	0.79	6.02*
25.	BC-0033	-12.93***	-24.93***	0.00	3.33	3.33	1.33	-2.27	-6.67***	-20.19***	-14.96***

Table 18. Dunnett's test performed on morphological trait means to examine differences between each of the 45 upland cotton genotypes and two checks (CB-14 and Mutant-1)

SL.	Genotype	PH		SBPP		DFF		BPP		BBP	
		CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1
26.	BC-0036	-9.80***	-21.80***	0.33	3.67	4.33*	2.33	-1.67	-6.07***	2.50	7.73***
27.	BC-0037	-17.20***	-29.20***	1.33	4.67*	3.0	1.0	-2.0	-6.40***	-1.46	3.77
28.	BC-0038	-16.07***	-28.07***	0.00	3.33	4.33*	2.33	-2.27	-6.67***	0.48	5.71*
29.	BC-0042	-23.20***	-35.20***	-1.33	2.0	3.67 .	1.67	-2.13	-6.53***	-24.08***	-18.85***
30.	BC-0045	0.47	-11.53***	-2.0	1.33	4.0*	2.0	-3.0	-7.40***	-6.66**	-1.43
31.	BC-0046	-1.40	-13.40***	-1.67	1.67	4.33*	2.33	-3.53	-7.93***	-12.96***	-7.73***
32.	BC-0047	-4.60*	-16.60***	0.67	4.0	3.33	1.33	-4.07 .	-8.47***	-0.25	4.98 .
33.	BC-0048	-5.60**	-17.60***	-5.0*	-1.67	5.0**	3.0	-3.47	-7.87***	-3.87	1.36
34.	BC-0049	-21.33***	-33.33***	-5.0*	-1.67	3.0	1.0	-2.80	-7.20***	-19.17***	-13.94***
35.	BC-0050	2.07	-9.93***	-3.0	0.33	4.0*	2.0	-2.47	-6.87***	-8.09***	-2.86
36.	BC-0052	0.13	-11.87***	-4.33 .	-1.0	3.67 .	1.67	-3.33	-7.73***	-2.42	2.81
37.	BC-0053	14.73***	2.73	-3.67	-0.33	3.33	1.33	-2.87	-7.27***	-29.99***	-24.76***
38.	BC-0054	2.93	-9.07***	-1.0	2.33	4.33*	2.33	-3.53	-7.93***	-24.32***	-19.09***
39.	BC-0055	1.13	-10.87***	-2.3	1.0	3.0	1.0	-1.20	-5.60**	-6.96**	-1.73
40.	BC-0057	25.33***	13.33***	-3.00	0.33	4.33*	2.33	-3.80	-8.20***	-32.17***	-26.94***
41.	BC-0058	42.93***	30.93***	0.33	3.67	5.0**	3.0	-2.60	-7.0***	-30.48***	-25.25***
42.	BC-0059	12.00***	0.00	-0.67	2.67	4.33*	2.33	-4.13 .	-8.53***	-12.25***	-7.02**
43.	BC-0060	-5.20**	-17.20***	-0.33	3.0	4.0*	2.0	-3.20	-7.60***	-21.68***	-16.45***
44.	BC-0061	-11.73***	-23.73***	1.0	4.33 .	5.0**	3.0	-3.20	-7.60***	-29.97***	-24.74***
45.	BC-0062	40.23***	28.23***	0.33	3.67	4.67**	2.67	-0.53	-4.93*	-31.17***	-25.94***

Here, significant codes denoted '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1; +, significant positive performance compared with two checks, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, BPP= bolls per plant and BBP= boll bursting percentage (%).

sometimes correlate with reduced boll development or compromised fiber quality (Zhao *et al.*, 2021).

Variation in sympodial branches and bolls per plant also presents a complex picture. Increases in branches and bolls, highlighted by TC-1903 and BC-0510, suggested higher yield potential, but these increases should be balanced against other factors like boll bursting percentage (BBP).

For example, genotype TC-1903 exhibited both a high number of bolls and a marked increase in boll bursting, the latter potentially reducing harvestable yield due to boll damage (Khan *et al.*, 2019). Conversely, genotypes with significantly reduced BBP, such as BC-0062 might have better boll retention but did not necessarily show corresponding improvements in bolls per plant, indicating that boll retention alone does not guarantee yield advantage.

Overall, while the observed significant differences underscored valuable genetic diversity, the practical breeding value of these genotypes depended on the integration of multiple traits and their interactions under field conditions. High-performing genotypes identified through this analysis warranted further multi-environment testing to confirm their stability and utility in breeding programs aiming for both yield and agronomic adaptability.

The **Table 19** represents the results of a Dunnett's test to compare the performance of key yield traits in 45 upland cotton genotypes against two standard check varieties, CB-14 and Mutant-1. The analysis provided a clear indication of which genotypes showed significant differences in key morphological and yield-related traits.

For single boll weight (SBW), several genotypes showed significantly increased weights compared to the checks. Genotypes BC-0027 displayed a highly significant increase of 1.04** g and 1.35*** g over CB-14 and Mutant-1, respectively. Genotype BC-0510 also showed a significant increase.

In seeds per boll (SPB), several genotypes exhibited superior performance. Genotype RA-9.N5, BC-0027 and TC-1903 showed an increase of 7.0, 4.7 and 3.55 compared to Mutant-1.

For seed cotton yield per plant (SCYPP), several genotypes showed superior performance. Genotypes BC-0491, BC-0027 and BC-0512 displayed highly significant

increases in SCYPP, with values of 10.50***, 10.36*** and 9.78***, respectively, compared to CB-14. Other genotypes, such as BC-0510 and RA-5 also exhibited highly significant increases compared to Mutant-1.

In terms of lint fiber yield per plant (LFYPP), a number of genotypes outperformed the checks. Genotypes RA-5, BC-0491, BC-0510, showed a highly significant increase of 13.06***, 12.83*** and 12.49*** compared to CB-14. Several other genotypes, including BC-0512, JA-13/X and BC-0027 also displayed highly significant increases.

Moreover, in the earliness index (EI), many genotypes were significantly earlier than the checks. The most notable differences were seen in genotypes BC-0510, BC-0512, BC-0491, RA-2 and RA-9.N5 which all showed a significant increase in their EI compared to both checks, indicating they are valuable genetic resources for incorporating earliness into new breeding lines. Conversely, other genotypes, like BC-0060, BC-0061 and BC-0062 were significantly later.

The **Table 19** highlights several genotypes with superior performance in traits such as single boll weight, seeds per boll, seed cotton yield, and lint fiber yield compared to the checks CB-14 and Mutant-1; these findings warranted cautious interpretation.

First, despite higher single boll weight and seed count, increased individual boll size or seed numbers do not always translate to an overall yield advantage under diverse field conditions. Larger bolls can sometimes be associated with delayed maturation or increased susceptibility to pests and diseases, potentially offsetting yield gains (Patel *et al.*, 2021). For instance, genotype BC-0027 showed significant increase in boll weight and yield, but without assessments of boll retention or fiber quality, the practical benefits remain uncertain. Similarly, genotypes like CB-2, CB-8 demonstrated high yield components but also showed delayed flowering and earliness index, suggesting a trade-off between yield potential and crop maturation rate. Late-maturing genotypes may be less adaptable to regions with short growing seasons or where early harvest is critical to avoid adverse weather (Singh & Kumar, 2020).

This highlighted the challenge in breeding programs to balance yield traits with crop phenology for target environments. Although some genotypes, such as BC-0024, BC-0028 and BC-0038, exhibited significant earliness, their lower performance in other yield components were evident in previous results, indicating that early maturity alone may not

Table 19. Dunnett's test performed on key yield contributing trait means to examine differences between each of the 45 upland cotton genotypes and two checks (CB-14 and Mutant-1)

SL.	Genotype	SBW		SPB		SCYPP		LFYPP		EI	
		CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1
1.	RA-2	0.49	0.80	-3.33	-7.33	7.75***	4.32 .	10.47***	4.45 .	7.73***	5.83***
2.	RA-5	0.58	0.89*	-4.0*	-1.40	9.40***	5.97**	13.06***	7.04***	6.31***	4.41***
3.	RA-9.N5	0.63	0.94*	-1.90	7.0	6.33***	2.90	8.88***	2.86	7.65***	5.75***
4.	CB-2	-0.37	-0.06	-4.03*	-1.43	3.07	-0.36	7.21***	1.19	-6.14***	-8.04***
5.	CB-6	0.07	0.38	-10.03***	-7.43***	4.79*	1.36	9.27***	3.25	2.00	0.10
6.	CB-7	-0.28	0.03	-6.43***	-3.83 .	0.39	-3.04	4.92*	-1.10	0.00	-1.90
7.	CB-8	-0.21	0.10	-5.90***	-3.30	3.32	0.11	7.26***	1.24	-1.24	-3.14
8.	CB-10	-0.26	0.05	-7.30***	-4.70**	0.53	2.90	4.64 .	-1.38	0.40	-1.50
9.	CB-14	-	0.31	-	2.60	-	3.43	-	-6.02**	-	-1.90
10.	BC-0491	0.59	0.90*	-3.97*	-1.37	10.50***	7.07***	12.83***	6.81***	7.98***	6.08***
11.	BC-0510	0.79	1.10**	-5.50**	-2.90	10.05***	6.12***	12.49***	6.47**	8.65***	6.75***
12.	BC-0512	0.58	0.89*	-3.67 .	-1.07	9.78***	6.35***	12.04***	6.02**	8.32***	6.41***
13.	JA-11/L	-0.35	-0.04	-8.97***	-6.37***	-0.93	-4.36 .	1.49	-4.53 .	2.76*	0.86
14.	JA-17/2	0.49	0.80 .	-3.53	-9.33	8.70***	5.27*	11.10***	5.08*	5.98***	4.08***
15.	JA-13/X	0.78 .	1.09**	-4.20*	-1.60	9.59***	6.16**	12.21***	6.19**	5.65***	3.75***
16.	Mutant-1	-0.31	-	-2.60	-	3.43	-	6.02**	-	1.90	-
17.	TC-1903	0.50	0.81 .	-2.60	3.55	8.58***	5.15*	11.55***	5.53*	4.98***	3.08*
18.	TC-1904	-0.12	0.19	-9.30	-6.70***	-9.08***	-12.51***	-5.96**	-11.98***	1.90	0.00
19.	BC-0024	-0.26	0.05	-11.63***	-9.03***	-9.64***	-13.07***	-6.69***	-12.71***	3.32**	1.41
20.	BC-0025	-0.32	-0.01	-6.63***	-4.03*	0.34	-3.09	2.57	-3.45	-0.24	-2.14
21.	BC-0026	0.01	0.32	-7.63***	-5.03**	-14.04***	-17.47***	-10.23***	-16.25***	2.77*	0.87
22.	BC-0027	1.04**	1.35***	-2.13	4.70	10.36***	6.93***	11.77***	5.74**	6.90***	5.0***
23.	BC-0028	-0.11	0.20	-8.63***	-6.03***	-1.30	-4.73*	2.05	-3.97	3.24**	1.33
24.	BC-0029	-0.35	-0.04	-9.34***	-6.74***	-9.46***	-12.89***	-6.79***	-12.81***	1.95	0.05
25.	BC-0033	-0.08	0.23	-9.30***	-6.70***	-12.04***	-15.47***	-9.83***	-15.85***	2.28	0.38

Table 19. Dunnett's test performed on key yield contributing trait means to examine differences between each of the 45 upland cotton genotypes and two checks (CB-14 and Mutant-1)

SL.	Genotype	SBW		SPB		SCYPP		LFYPP		EI	
		CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1	CB-14	M-1
26.	BC-0036	-0.09	0.22	-11.63***	-9.03***	-8.98***	-12.41***	-6.53***	-12.55***	3.0*	1.10
27.	BC-0037	-0.35	-0.04	-12.23***	-9.63***	-8.85***	-12.28***	-6.43**	-12.45***	3.10*	1.20
28.	BC-0038	-0.49	-0.18	-8.97***	-6.37***	-6.12**	-9.55***	-2.22	-8.24***	3.20**	1.30
29.	BC-0042	-0.18	0.12	-8.43***	-5.83***	2.25	-1.18	5.36*	-0.66	1.77	-0.13
30.	BC-0045	-0.31	0.003	-8.63***	-6.03***	-0.11	-3.54	1.94	-4.08	1.88	-0.02
31.	BC-0046	-0.16	0.15	-11.30***	-8.70***	-9.58***	-13.01***	-6.90***	-12.92***	2.88*	0.98
32.	BC-0047	-0.15	0.16	-10.63***	-8.03***	-2.12	-5.55**	0.53	-5.49*	0.77	-1.13
33.	BC-0048	-0.25	0.06	-7.76***	-5.16**	-2.34	-5.77**	1.82	-4.20	-3.10*	-5.00***
34.	BC-0049	-0.17	0.14	-9.63***	-7.03***	-14.52***	-17.95***	-11.24***	-17.26***	1.90	0.0
35.	BC-0050	-0.41	-0.10	-9.93***	-7.33***	-10.33***	-13.76***	-6.25**	-12.27***	-0.12	-2.02
36.	BC-0052	-0.54	-0.23	-6.63***	-4.03*	-9.19***	-12.62***	-6.75***	-12.77***	-2.10	-4.00***
37.	BC-0053	0.16	0.47	-9.93***	-7.33***	-13.78***	-17.21***	-10.24***	-16.26***	-1.35	-3.25**
38.	BC-0054	-0.04	0.27	-10.63***	-8.03***	-11.33***	-14.76***	-8.08***	-14.10***	-2.23	-4.13***
39.	BC-0055	-0.13	0.18	-12.30***	-9.70***	-7.96***	-11.39***	-5.57**	-11.59***	-0.12	-2.02
40.	BC-0057	0.07	0.38	-11.63***	-9.03***	-15.71***	-19.14***	-12.45***	-18.47***	1.77	-0.13
41.	BC-0058	-0.51	-0.20	-13.23***	-1.06***	-9.80***	-13.23***	-6.86***	-12.88***	-0.10	-2.0
42.	BC-0059	-0.07	0.24	-13.63***	-1.10***	-11.90***	-15.33***	-8.67***	-14.69***	0.88	-1.02
43.	BC-0060	-0.09	0.22	-10.97***	-8.37***	-1.84	-5.27*	0.82	-5.20*	-9.79***	-11.69***
44.	BC-0061	-0.41	-0.10	-12.30***	-9.70***	-11.25***	-14.68***	-8.45***	-14.47***	-9.23***	-11.13***
45.	BC-0062	-0.26	0.05	-10.63***	-8.03***	-1.63	-5.06*	1.02	-5.00*	-7.25***	-9.15***

Here, significant codes denoted '***' 0.001 '**' 0.01 '*' 0.05 '.' 0.1 ' ' 1; +, significant positive performance compared with two checks, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g) and EI= earliness index (%)

guarantee higher productivity. This underscores the need for multi-trait selection and evaluation under different agronomic conditions to identify genotypes with stable and balanced performance.

Several studies in crop science have employed Dunnett's test as a statistical method for comparing multiple genotypes or treatments with a designated control. For instance, Xu *et al.* (2020) utilized Dunnett's multiple range test to assess the effects of salt priming on rice (*Oryza sativa*) seedlings a methodological approach analogous to that adopted in the present study. Similarly, Goldwasser *et al.* (2012) implemented a two-sided Dunnett's test to evaluate differences among chickpea (*Cicer arietinum*) genotypes in their resistance to *Cuscuta* infection.

4.2.4 Cluster analysis among 45 upland cotton genotypes

Table 20 summarized the hierarchical clustering analysis that classified 45 upland cotton genotypes into three distinct clusters based on their morphological traits. Cluster I was the smallest group with 10 genotypes, cluster III was the largest with 18 genotypes, and cluster II contained 17 genotypes (**Figure 9**). The formation of these distinct clusters indicated genetic dissimilarity among genotypes, with genotypes within the same cluster being more genetically similar to each other than to those in other clusters. The hierarchical clustering analysis revealed three distinct clusters among the cotton genotypes, underscored the presence of substantial genetic diversity within the population. Such genetic differentiation is essential for practical breeding, as it allows breeders to identify genetically divergent parents that can be exploited to produce heterotic hybrids with improved performance (Dabholkar, 1992).

The smaller size of cluster I compared to clusters II and III suggested a more uniform genetic makeup within that group, whereas larger clusters might contain greater intra-cluster variability. Understanding this structure aids in strategic parent selection, as crossing genotypes from genetically distant clusters generally leads to higher heterosis and a broader genetic base in progeny (Dabholkar, 1992). These findings aligned with established principles that advocate utilizing genetic diversity revealed by cluster analysis to enhance breeding efficiency and crop improvement.

Table 20. Cluster groups of 45 cotton genotypes based on hierarchical clustering

Cluster	Genotype position	Number of genotypes	Genotypes included
I	1, 2, 3, 10, 11, 12, 14, 15, 17, 22	10	RA-2, RA-5, RA-9.N5, BC-0491, BC-0510, BC-0512, JA-17/2, JA-13/X, TC-1903, BC-0027
II	4, 5, 6, 7, 8, 9, 13, 16, 20, 23, 26, 27, 28, 29, 30, 32, 33	17	CB-2, CB-6, CB-7, CB-8, CB-10, CB-14, JA-11/L, Mutant-1, BC-0025, BC-0028, BC-0036, BC-0037, BC-0038, BC-0042, BC-0045, BC-0047, BC-0048
III	18, 19, 21, 24, 25, 31, 34, 35, 36, 37, 38, 39, 40, 41, 42, 43, 44, 45	18	TC-1904, BC-0024, BC-0026, BC-0029, BC-0033, BC-0046, BC-0049, BC-0050, BC-0052, BC-0053, BC-0054, BC-0055, BC-0057, BC-0058, BC-0059, BC-0060, BC-0061, BC-0062

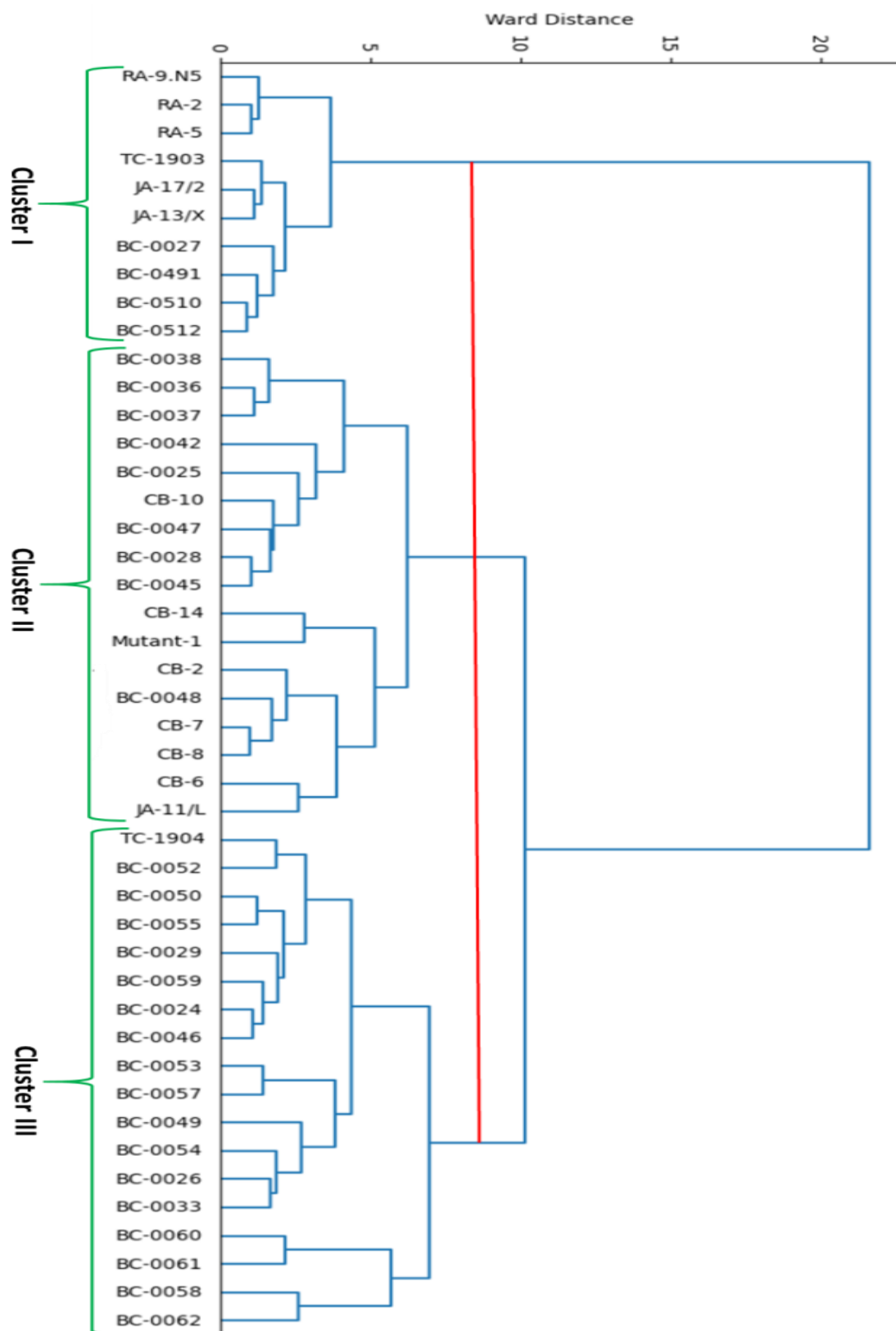


Figure 9. UPGMA clustering of 10 morphological parameters in 45 upland cotton genotypes

The **Table 21** represents the mean values of ten morphological traits for the three identified clusters, providing a detailed characterization of each group. Cluster mean analysis is a crucial follow-up to clustering because it helps breeders to discern the traits that define each cluster and identify the most promising groups for selection and breeding (Dabholkar, 1992).

The cluster I stood out with the highest mean values for key yield-related traits, including sympodial branches per plant (SBPP), bolls per plant (BPP), boll bursting percentage (BBP), single boll weight (SBW), seeds per boll (SPB), seed cotton yield per plant (SCYPP), lint fiber yield per plant (LFYPP) and earliness index (EI).

The mean SCYPP in cluster I was substantially greater than those of cluster II (116.46 g) and cluster III (107.03 g), underscoring its superior productivity and desirability for yield improvement. This suggested that genotypes in cluster I possessed advantageous gene combinations favorable for enhancing cotton yield (Dabholkar, 1992).

The cluster II exhibited intermediate performance across most traits (**Table 21**). Although the mean SCYPP and LFYPP values for cluster II fallen below those of cluster I, they surpassed those of cluster III. This cluster might provide a valuable balance of traits and serve as a source of parental lines for crossing with genotypes from other clusters to generate new genetic variations.

In contrast, cluster III showed the lowest mean values for most important yield-associated traits, such as SCYPP (107.03 g), LFYPP (93.39 g), BBP (68.23%), and SBW (4.58 g). Genotypes in this cluster typically demonstrated lower productivity and might be better suited for breeding focused on traits other than yield, such as disease resistance (Falconer & Mackay, 1996). Notably, this cluster exhibited the highest mean plant height (PH), reinforcing the previously reported negative correlation between plant height and yield.

The **Table 22** displayed the average intra-cluster and inter-cluster genetic distances among the three cotton genotype clusters. The diagonal, bolded values represented intra-cluster distances, reflecting the genetic variability within each cluster. Off-diagonal values represented inter-cluster distances, indicating genetic divergence between clusters.

The intra-cluster distances for clusters I, II, and III were 1.15, 1.95, and 1.97, respectively. This variable intra-cluster diversity could inform breeding strategies based on whether uniformity or variability is sought within a parental group (Singh & Narayanan, 1993).

Table 21. Cluster mean of 10 morphological parameters in 45 upland cotton genotypes under three clusters

Cluster	Characters									
	PH (cm)	SBPP	DFF	BPP	BBP	SBW (g)	SPB	SCYPP (g)	LFYPP (g)	EI (%)
I	125.25 (L)	22.23 (H)	52.50 (L)	29.55 (H)	94.16 (H)	5.42 (H)	30.15 (H)	126.31 (H)	112.21 (H)	93.37 (H)
II	130.06	17.96 (L)	59.69	24.42	83.64	4.54 (L)	26.06	116.46	102.91	87.13
III	140.46 (H)	18.18	61.00 (H)	23.47 (L)	68.23 (L)	4.58	23.08 (L)	107.03 (L)	93.39 (L)	85.65 (L)

Here, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, BPP= bolls per plant, BBP= boll bursting percentage (%), SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness Index (%), H = high and L = low.

Table 22. Intra (bold and diagonal) and inter-cluster distances of 45 upland cotton genotypes

Cluster	I	II	III
I	1.15	5.19	6.77
II		1.95	3.66
III			1.97

The cluster I exhibited the lowest intra-cluster distance, suggesting that genotypes within this cluster are more genetically uniform. Conversely, the highest genetic variability within a cluster was found in cluster III. Inter-cluster distances exceeded intra-cluster distances for all pairs, underscoring the distinctiveness of the three clusters. The most significant genetic divergence was observed between clusters I and III (6.77), indicating extensive genetic differentiation, followed by clusters I and II (5.19), with the smallest distance between clusters II and III (3.66) which were more closely related (**Table 22**).

This pattern provided a valuable guide for selecting genetically divergent parents to maximize heterosis and broaden the genetic base in breeding materials. Crosses between genotypes from clusters with larger genetic distances, such as clusters I and III, were expected to yield superior hybrids due to increase heterozygosity and segregation variance (Falconer & Mackay, 1996). Based on the results of genetic diversity and earliness index, the genotypes from cluster II might be used as parent in hybridization program for the development of early boll picking and high yielding cotton variety (Akter *et al.*, 2019).

4.2.5 Principal component analysis

The principal component analysis (PCA) resulted for ten morphological traits of 45 upland cotton genotypes were shown in **Table 23**. The table presented loading values, which reflected each trait's correlation with the corresponding principal component (PC), as well as eigenvalues, percentage of variance explained, and cumulative variance. Although ten PCs were extracted, **Figure 10** illustrated that the first three PCs accounted for the majority of the total variance (>82.27%). PC1 had the largest eigen value, explaining 61.43% of the variance, followed by PC2 and PC3 explaining 11.37% and 9.48%, respectively. Together, these three components captured over 82.27% of the total variation, facilitating more focused selection and breeding efforts (Singh & Narayanan, 1993).

PC1 was chiefly defined by high positive loadings for bolls per plant (BPP, 0.867), boll bursting percentage (BBP, 0.770), single boll weight (SBW, 0.833), seeds per boll (SPB, 0.814), seed cotton yield per plant (SCYPP, 0.898), lint fiber yield per plant (LFYPP, 0.879) and earliness index (EI, 0.724) (**Table 23**). This highlighted the collective importance of these traits as primary drivers of genetic diversity and productivity in cotton, reinforcing their value as targeted for breeding programs aimed at yield improvement (Falconer & Mackay, 1996). Traits with strong positive loadings on a PC are highly

Table 23. Arrangement of principal components against 11 morphological characteristics in 45 upland cotton genotypes

Character	PC1	PC2	PC3	PC4	PC5
Plant height (cm)	-0.418	0.498	0.619	0.418	0.118
Sympodial branches per plant	0.584	0.640	-0.146	-0.350	0.274
Days to 50% flowering	-0.907	-0.050	0.095	-0.138	0.110
Bolls per plant	0.867	0.305	0.104	-0.042	-0.106
Boll bursting percentage (%)	0.770	-0.434	-0.047	0.170	0.356
Single boll weight (g)	0.833	0.291	-0.157	0.038	-0.262
Seeds per boll	0.814	-0.207	0.286	0.050	-0.268
Seed cotton yield per plant (g)	0.898	-0.144	0.329	-0.141	0.095
Lint fiber yield per plant (g)	0.879	-0.186	0.338	-0.134	0.097
Earliness index (%)	0.724	0.108	-0.439	0.458	0.101
Eigen value	6.143	1.137	0.948	0.599	0.408
Percentage of variance (%)	61.426	11.366	9.477	5.991	4.083
Cumulative percentage (%)	61.426	72.793	82.269	88.261	92.343

Here, PC= principal component

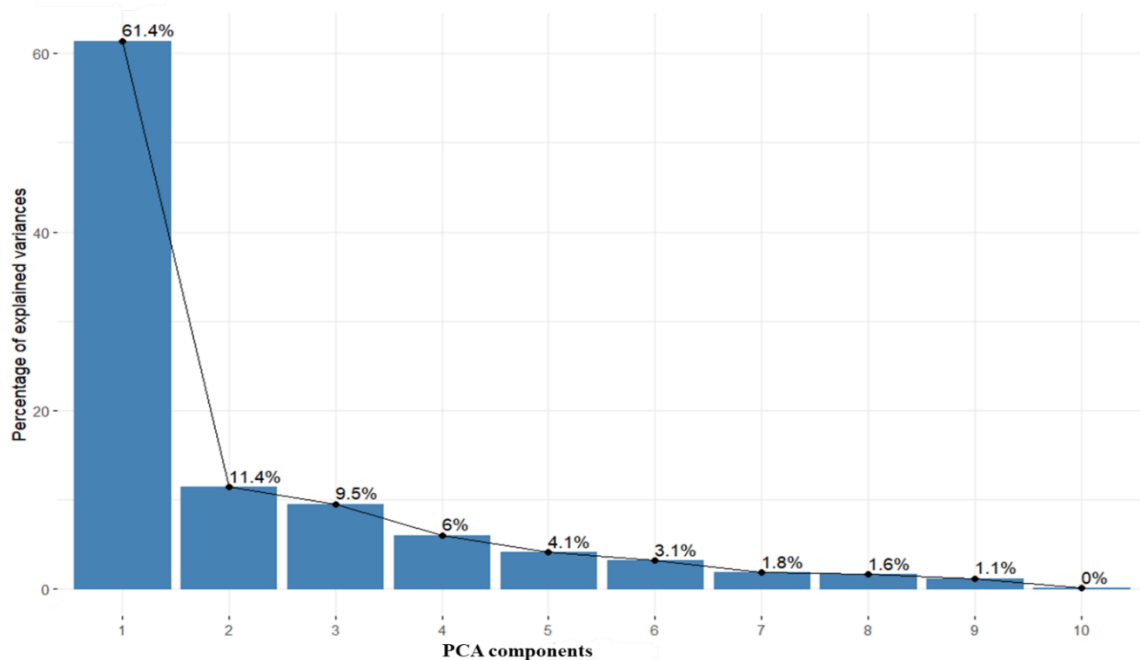


Figure 10. Scree plots of 10 morphological parameters in 45 upland cotton genotypes

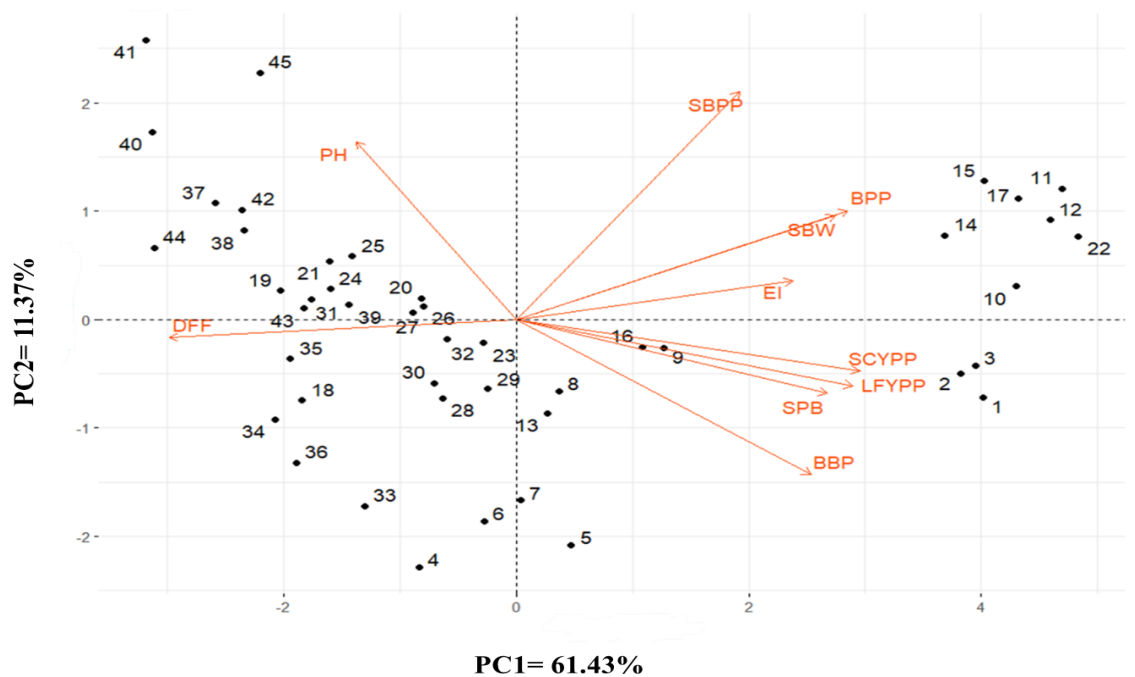


Figure 11. PCA biplot of 10 morphological parameters in 45 upland cotton genotypes

Here, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, BPP= bolls per plant, BBP= boll bursting percentage (%), SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness index (%), PC= principal component, and the numbers showing genotypes.

correlated with that component. PC2 was primarily associated with plant height (PH, 0.498), sympodial branches per plant (SBPP, 0.640) and bolls per plant (BPP, 0.305), while PC3 also reflected plant height (0.619), seed cotton yield per plant (SCYPP, 0.329) and lint fiber yield per plant (LFYPP, 0.338) with differing loadings (**Table 23**) suggesting these morphological traits contributed to variability independent from yield components. This distinction underscored the multidimensional nature of genetic variation and the need to consider both yield and morphological traits when selecting genotypes for enhancement. The grouping of genotypes possessing excelled traits signifies genetic potential of the germplasm for the improvement of seed cotton yield characteristics in cotton crop (Liaqat *et al.*, 2015).

4.2.6 Biplot analysis of 45 upland cotton genotypes

The **Figure 11** displayed the principal component analysis (PCA) biplot, visually summarized the relationships among ten morphological traits and 45 upland cotton genotypes. The biplot presents the first two principal components (PC1 and PC2) on the X- and Y-axes, respectively. PC1 accounted for (61.4%) of the total variance, while PC2 explained 11.40%. Traits are represented by vectors, with their length and direction indicating the degree and nature of their contribution to the components. Genotypes are shown as points, and their proximity to trait vectors reflected their performance for those traits.

The biplot revealed that vector for seed cotton yield per plant (SCYPP), lint fiber yield per plant (LFYPP), seeds per boll (SPB) and boll bursting percentage (BBP) pointed in similar directions, illustrating strong positive correlations among these traits (**Figure 11**). Their relatively long vectors indicated substantial contributions to the overall variation (Singh & Narayanan, 1993). Genotypes RA-2 (1), RA-5 (2), and RA-9.N5 (3) were clustered near these vectors, suggesting high-yield potential.

Conversely, the days to 50% flowering (DFF) vector pointed in the opposite direction, signifying a negative correlation with yield traits emphasizing the advantage of earlier flowering genotypes in breeding. Vectors for plant height (PH), sympodial branches per plant (SBPP), and boll bursting percentage (BBP) occupied different quadrants, highlighting their distinct influence in phenotypic diversity. Genotypes BC-0058 (41), BC-0057 (40), and BC-0062 (45) were positioned distant from the yield-related vectors, indicating lower yield potential (Falconer & Mackay, 1996).

4.3 Experiment III: Assessment of fiber quality and molecular profiling of early boll picking well performing 12 upland cotton (*Gossypium hirsutum* L.) genotypes

With a view of reaching final selection of early boll harvesting, higher seed cotton yield and quality fiber producing cotton genotypes, apparently genetically diverged 12 genotypes including two checks (**Table 3**) were selected from 45 genotypes considering individual performances measured by different biometrical tools and then the selected 12 genotypes were evaluated under field conditions and recorded data were analysed for morphological and qualitative traits of fiber, and molecular characterization was performed from DNA samples. However, all analyses with results are discussed below-

4.3.1 Analysis of variance (ANOVA) of morphological characters in early boll picking well performing 12 upland cotton genotypes

The analysis of variance (ANOVA) for eleven morphological traits in twelve upland cotton genotypes (**Table 24**) revealed significant variability among genotypes for all traits studied. Highly significant ($p \leq 0.001$) differences were recorded for plant height, sympodial branches per plant, days to 50% flowering, days to first boll splitting, seed cotton yield per plant, lint fiber yield per plant, and earliness index, while bolls per plant and single boll weight were significant at the 1% probability level. Seeds per boll showed significance at the 5% level. The presence of such highly significant differences indicated a broad genetic base and sufficient variability among the genotypes, which was a prerequisite for successful selection in cotton improvement programs.

The coefficient of variation (CV%) ranged from 0.86% for days to first boll splitting to 6.85% for bolls per plant, reflecting high experimental precision. The mean performance and range of values further confirmed the diversity among the genotypes, with plant height varying from 116.62 to 144.08 cm (mean 130.83 ± 1.267 cm), sympodial branches from 15.33 to 22.47 (mean 18.90 ± 0.325), and seed cotton yield per plant from 114.04 to 126.83 g (mean 122.60 ± 0.714 g) (**Table 24**). Similar ranges of variation had been documented in earlier studies, supporting the reliability of the present findings. The results demonstrated that traits like plant height, yield components, and earliness contributed substantially to total variability and might be targeted for genetic improvement. These findings were consistent with Ahmad *et al.* (2008), who reported highly significant genotypic differences for key morphological traits associated with earliness and yield.

Table 24. Mean squares (MS) derived on 10 morphological characters in well performing 12 upland cotton genotypes

Traits	Mean Sum of Square					
	Genotype	Replication	Error	CV%	Mean Range	Grand Mean $\pm$ SE
Plant height (cm)	162.27***	1.98	4.82	1.68	116.62-144.08	130.83 $\pm$ 1.27
Sympodial branches per plant	6.043***	0.89	0.32	2.98	15.33-22.47	18.90 $\pm$ 0.33
Days to 50% flowering	17.80***	0.50	1.10	2.04	47.10-57.93	51.49 $\pm$ 0.61
Days to first boll splitting	76.75***	0.28	0.79	0.86	97.35-118.10	103.50 $\pm$ 0.51
Bolls per plant	15.90**	1.09	3.81	6.85	23.0-33.70	28.47 $\pm$ 1.13
Single boll weight (g)	0.36**	0.01	0.08	5.77	4.05-5.69	4.90 $\pm$ 0.16
Seeds per boll	10.11*	0.63	4.10	6.82	24.20-33.73	29.66 $\pm$ 1.17
Seed cotton yield per plant (g)	26.91***	2.86	1.53	1.01	114.04-126.83	122.60 $\pm$ 0.71
Lint fiber yield per plant (g)	36.30***	3.96	2.50	1.48	97.46-112.96	106.88 $\pm$ 0.91
Earliness index (%)	11.06***	0.03	1.23	1.22	86.58-94.24	90.50 $\pm$ 0.64

Here, ***, ** and * indicated significant at 0.1%, 1% and 5% levels of probability, respectively, CV= co-efficient of variation and SE= standard error of mean.

Abro *et al.* (2021) found significant variation across genotypes and F₄ populations for morphological, yield, and fiber traits.

4.3.2 Mean performance of the morphological traits in early boll picking well performing 12 upland cotton genotypes

The selected 12 lines were analyzed in 2023 and the mean performance are presented in **Table 25**. The mean performance revealed that the differences among the twelve upland cotton genotypes were statistically significant for all eleven morphological traits studied, suggesting the presence of considerable genetic variability.

Plant height showed a wide range, from 118.53 cm to 142.38 cm, with the tallest plants recorded in Mutant-1, while BC-0510 and BC-0512 produced comparatively shorter plants.

The number of sympodial branches per plant varied from 15.93 to 21.26, with TC-1903 producing the highest number, indicating better fruiting potential. Variation was also observed in flowering and maturity traits. Days to 50% flowering ranged from 48.50 days in RA-2 to 56.67 days in Mutant-1, while days to first boll splitting ranged from 97.70 days in BC-0491 to 116.63 days in CB-14. This indicated that RA-2 and BC-0491 were earlier in maturity, whereas Mutant-1 and CB-14 were relatively late maturing.

Yield components also showed marked differences. The number of bolls per plant ranged from 24.58 in CB-14 to 31.95 in RA-2, and single boll weight varied from 4.18 g in Mutant-1 to 5.27 g in BC-0512. Seeds per boll were highest in BC-0491 (32.28) and lowest in Mutant-1 (26.04). Seed cotton yield per plant ranged from 116.35 g to 125.88 g, with RA-2, BC-0510, and BC-0512 showing superior performance. Lint fiber yield followed a similar pattern, with BC-0510 (111.91 g), BC-0512 (110.44) and RA-2 (110.03 g) leading, while Mutant-1 (99.43 g) was the lowest (**Table 25**).

Earliness index values varied from 87.65% in Mutant-1 to 93.27% in RA-2, indicating that RA-2 combined high yield with early maturity. Overall, RA-2, BC-0510, and BC-0512 were identified as promising genotypes due to their desirable combination of yield and earliness, while Mutant-1 was characterized by tall stature, late maturity, and comparatively lower yield performance.

Table 25. Mean performance of 10 morphological characteristics in well performing 12 upland cotton genotypes

Genotype	PH (cm)	SBPP	DFF	DFBS	BPP	SBW (g)	SPB	SCYPP (g)	LFYPP (g)	EI (%)
RA-2	132.75 cd	17.99 de	48.50 d	102.98 c	31.95 a	4.99 a	30.51 a	125.24 a	110.03 ab	93.27 a
RA-5	132.34 cd	18.06 de	51.32 b	102.63 c	31.18 ab	5.12 a	30.18 a	121.39 d	107.22 c-e	90.59 b-d
RA-9.N5	130.91 d	17.68 e	50.36 bc	100.07 d	28.03 b-d	5.05 a	30.02 a	122.17 cd	106.02 de	89.87 c-e
BC-0491	123.91 e	18.8 cd	51.81 b	97.70 e	26.52 c-e	5.15 a	32.28 a	123.94 a-c	108.16 b-d	91.84 ab
BC-0510	118.98 f	19.92 b	48.79 cd	100.07 d	30.81 ab	4.94 a	29.99 a	125.88 a	111.91 a	91.63 a-c
BC-0512	118.53 f	19.37 bc	50.56 bc	103.67 c	28.66 a-c	5.27 a	29.28 ab	124.55 ab	110.44 ab	92.21 ab
JA-17/2	136.49 b	19.53 bc	50.49 bc	103.02 c	28.08 b-d	4.98 a	30.47 a	124.16 a-c	105.50 de	90.81 b-d
JA-13/X	137.35 b	19.99 b	51.47 b	99.55 d	28.14 b-d	5.01 a	30.18 a	123.11 b-d	107.00 c-e	88.54 ef
TC-1903	134.41 b-d	21.26 a	50.78 b	102.63 c	29.54 a-c	4.95 a	29.43 ab	122.64 b-d	105.41 e	89.18 d-f
BC-0027	127.16 e	20.09 b	51.35 b	103.72 c	29.13 a-c	4.93 a	31.34 a	124.43 ab	108.91 bc	92.72 a
CB-14	134.72 bc	18.22 de	55.78 a	116.63 a	24.58 de	4.19 b	26.20 b	117.26 e	102.52 f	87.71 f
Mutant-1	142.38 a	15.93 f	56.67 a	109.33 b	24.96 e	4.18 b	26.04 b	116.35 e	99.43 g	87.65 f
SD	7.36	1.47	2.51	4.96	2.73	4.41	2.41	3.10	3.63	2.06
LSD (5%)	3.72	0.95	1.78	1.50	3.30	0.478	3.43	2.10	2.68	1.88

Here, the mean values bearing same letter (s) didn't differ significantly at $p < 0.05$, PH= plant height (cm), SBPP= sympodial branches per plant, DFF= days to 50% flowering, DFBS= days to first boll splitting, BPP= bolls per plant, SBW= single boll weight (g), SPB= seeds per boll, SCYPP= seed cotton yield per plant (g), LFYPP= lint fiber yield per plant (g), EI= earliness Index (%), SD= standard deviation and LSD (5%) = least significance difference at 5% level of probability.

plant height (PH), bolls per plant (BPP), and seed cotton yield per plant (SCYPP), similar to the higher values observed in RA-2, BC-0510, and BC-0027. Ahsan *et al.* (2015) observed that late-maturing genotypes produced lower yields, reflecting the lower seed cotton yield per plant (SCYPP), lint fiber yield per plant (LFYPP), and earliness index (EI) in Mutant-1 and CB-14.

4.3.3 Analysis of variance (ANOVA) of fiber quality characters of early boll picking well performing 12 cotton genotypes

The analysis of variance (ANOVA) for twelve fiber quality traits in twelve upland cotton genotypes revealed highly significant differences ($p < 0.001$) among genotypes for most of the studied traits, except ginning out turn (GOT), which showed significance at the 5% level (**Table 26**). This indicated the presence of considerable genetic variability that could be exploited through selection of breeding programs.

The traits related to fiber length, such as upper half mean length (UHML) and mean length, exhibited highly significant variation and ranged between 27.45–31.22 mm with a grand mean of 29.35 mm, while mean length varied from 22.33–26.55 mm with an average of 24.43 mm. These results confirmed the diversity in fiber elongation potential among genotypes, which was consistent with earlier findings that emphasized the importance of fiber length as a key determinant of spinning performance (Khan *et al.*, 2020; Smith & Jones, 2019). Bano *et al.*, 2023 opined that the results of ANOVA revealed that all the genotypes were highly significant for most of the traits like seed cotton yield per plant, lint index, lint percentage, seed index, seed density, seed volume, fiber fineness, fiber strength and fiber length.

Similarly, significant variation was observed for uniformity index (81.35–85.04%), short fiber index (7.1–10.0%), and strength (28.5–29.3 g/tex), with low coefficients of variation (CV%), highlighting the precision of measurement and stability across replications (**Table 26**). The low error variance further supported the reliability of these estimates. Previous studies also reported substantial variation in fiber strength and uniformity index across cotton genotypes, suggesting their potential utility in developing high-quality cultivars (Zhang *et al.*, 2019).

Fiber fineness traits such as micronaire (4.46–5.47) and maturity ratio (0.86–0.89) also showed significant differences. The narrow range for maturity ratio indicated relative

Table 26. Mean squares (MS) derived on 12 fiber quality characters in well performing 12 upland cotton genotypes

Traits	Mean Sum of Square					
	Genotype	Replication	Error	CV%	Mean Range	Grand Mean $\pm$ SE
Upper half mean length	2.894***	0.001	0.001	0.01	27.45-31.22	29.35 $\pm$ 0.002
Mean length	3.596***	0.000	0.001	0.02	22.33-26.55	24.43 $\pm$ 0.002
Uniformity index	2.688***	0.001	0.001	0.01	81.35-85.04	83.22 $\pm$ 0.003
Short fiber index	1.857***	0.001	0.001	0.03	7.10-10.00	8.34 $\pm$ 0.002
Strength	0.234***	0.001	0.001	0.00	28.50-29.30	28.84 $\pm$ 0.00
Elongation	0.154***	0.000	0.001	0.00	2.31-2.98	2.59 $\pm$ 0.00
Micronaire	0.341***	0.001	0.001	0.08	4.46-5.47	5.08 $\pm$ 0.002
Maturity ratio	0.004***	0.001	0.001	0.74	0.86-0.89	0.88 $\pm$ 0.004
Moisture	3.269***	0.000	0.000	0.08	3.50-6.46	5.13 $\pm$ 0.002
Reflectance	17.912***	0.000	0.000	0.01	65.50-72.70	70.12 $\pm$ 0.002
Yellowness	0.188***	0.001	0.000	0.05	5.80-6.50	6.19 $\pm$ 0.002
Ginning out turn	4.511*	0.315	1.485	1.40	84.98-88.90	87.17 $\pm$ 0.70

Here, CV= co-efficient of variation and SE= standard error of mean

uniformity, while micronaire variation suggested differential adaptability of genotypes to microclimatic and physiological conditions. These findings were in agreement with reports that micronaire is often influenced both genetically and environmentally (Patil *et al.*, 2021).

Moisture content and fiber reflectance (Rd) showed large differences among genotypes. Reflectance values ranged from 65.5–72.7, with an overall mean of 70.12, indicating noticeable variability in fiber brightness. This variability had direct implication for textile processing and market preferences (Liu *et al.*, 2017). Yellowness (+b) also varied significantly (5.8–6.5), although within a narrow range, highlighting differences in fiber pigmentation that could influence overall fiber grade.

The ginning out turn (GOT) ranged from 84.98-88.90%, with a mean of 87.17%. Unlike other traits, and exhibited significance at the 5% level (**Table 26**), suggesting relatively less genetic variability among the tested genotypes. This was in line with earlier findings where GOT exhibited narrower genetic variation compared to fiber quality traits (Rauf *et al.*, 2019). Nevertheless, even slight differences in GOT are economically important, as higher lint yield directly contributes to overall cotton productivity.

Overall, the highly significant differences among genotypes for most traits highlighted the presence of exploitable genetic variation in the studied materials. Traits such as fiber length, uniformity index, and reflectance exhibited particularly strong variability, making them promising targets for selection. The results corroborated previous studies emphasizing that multi-trait variability analysis is crucial in breeding programs aimed at developing superior upland cotton varieties with improved lint yield and fiber quality (Ali *et al.*, 2019; Zhang *et al.*, 2019).

4.3.4. Mean performances of selected cotton genotypes based on fiber quality parameters

Twelve fiber quality traits were evaluated across the selected 12 well performing early boll picking upland cotton genotypes (**Table 27**). The mean performance supported the findings of variance analysis (**Table 26**), while also illustrating the degree of variability and reliability associated with each trait.

Upper Half Mean Length (UHML) and Mean Length (ML) ranged significantly from 27.45 mm to 31.22 mm and 22.33 mm to 26.55 mm, respectively. Genotype BC-0491

showed the longest fibers (UHML = 31.22 mm, ML = 26.55 mm), while JA-17/2 recorded the shortest. These traits are essential for fiber spinning efficiency and yarn strength; as longer fibers contribute to fewer breaks during processing. Fiber length is a vital trait for yarn strength and spinning performance, and the observed variation reflected the genetic diversity within the studied materials (Khan *et al.*, 2020; Smith & Jones, 2019).

The Uniformity Index (UI) was highest in BC-0491 (85.04%) and lowest in JA-17/2 (81.35%), reflecting significant genotypic differences in fiber consistency. Low SFI values are desirable, with BC-0491 (7.1%) and JA-13/X (7.7%) demonstrating superior fiber integrity. Conversely, JA-17/2 exhibited the highest SFI (10.1%), indicative of more short fibers that can impair yarn smoothness (**Table 27**). These results aligned with earlier studies where genotypes with superior uniformity exhibited lower proportions of short fibers (Zhang *et al.*, 2019).

All genotypes had statistically similar fiber strength (Str.), ranging narrowly between 28.5–29.3 g/tex, indicating strong tensile performance across environments. This supported previous findings that fiber strength is predominantly under genetic control and less prone to environmental fluctuations (Ali *et al.*, 2019). Similarly, elongation (Elong.) showed no significant differences, with values around 2.31–2.98%, which falls within acceptable commercial limits.

Micronaire (Mic.), representing fiber fineness and maturity, varied significantly from 4.46 µg/inch (RA-2) to 5.47 µg/inch (BC-0491), with most values falling within the premium commercial range (3.7–4.9), though BC-0491 and CB-14 were slightly higher (**Table 27**). Maturity Ratio (MR) ranged from 0.86–0.89, classifying most fibers as mature based on cotton incorporated guidelines (MR > 0.8). These findings agreed with Patil *et al.* (2021), who reported narrow genetic variability for maturity ratio in upland cotton. These results confirmed that micronaire is more environmentally sensitive; whereas MR is relatively stable (Patil *et al.*, 2021).

Moisture ranged from 3.5% (JA-13/X) to 6.46% (CB-14), with significant differences among genotypes. Low moisture content is advantageous for lint processing and storage stability, while higher levels can increase susceptibility to microbial degradation. Reflectance (Rd), which indicates brightness, ranged from 65.5 (CB-14) to 72.7 (JA-13/X), while yellowness (+b) values ranged between 5.8–6.5. Fiber reflectance directly

Table 27. Mean comparison of 12 fiber quality parameters of well performing 12 upland cotton genotypes

SL	Genotype	UHML (mm)	ML (mm)	UI (%)	SFI	Str. (g/tex)	Elong. (%)	Mic. (µg/inch)	MR	Moist. (%)	Rd	+b	CG	GOT (%)
1.	RA-2	29.66 e	24.77 e	83.51 e	8.00 h	28.50 a	2.31 a	4.46 k	0.86 c	5.89 d	70.90 d	6.20 d	51-1	87.85 ^{a-c}
2.	RA-5	28.70 j	23.71 j	82.61 j	8.80 c	29.20 a	2.85 a	5.41 c	0.89 a	5.81 e	69.90 f	6.30 c	51-2	88.32 ^{ab}
3.	RA-9.N5	29.49 f	24.58 f	83.35 f	8.10 g	28.60 a	2.41 a	4.63 j	0.86 c	5.94 c	71.70 c	5.90 f	51-1	86.78 ^{b-e}
4.	BC-0491	31.22 a	26.55 a	85.04 a	7.10 k	29.30 a	2.98 a	5.47 a	0.89 a	5.47 f	68.80 g	6.50 a	51-2	87.27 ^{a-d}
5.	BC-0510	30.18 b	25.35 b	84.00 b	7.70 j	28.75 a	2.56 a	4.81 h	0.87 bc	4.88 h	72.40 b	6.10 e	51-1	88.90 a
6.	BC-0512	28.88 h	23.91 h	82.79 h	8.60 e	29.05 a	2.75 a	5.28 d	0.89 a	4.18 i	65.90 h	6.50 a	61-1	88.67 ^{ab}
7.	JA-17/2	27.45 l	22.33 l	81.35 l	10.1a	29.05 a	2.75 a	5.28 d	0.89 a	3.98 j	72.40 b	5.90 f	51-1	84.98 e
8.	JA-13/X	30.16 c	25.32 c	83.95 c	7.70 j	28.70 a	2.51 a	4.80 i	0.87 bc	3.50 l	72.70 a	5.80 g	51-1	86.92 ^{a-e}
9.	TC-1903	28.81 i	23.83 i	82.71 i	8.70 d	28.90 a	2.66 a	5.08 f	0.88 ab	3.80 k	71.70 c	6.10 e	51-1	85.95 ^{c-e}
10.	BC-0027	29.84 d	24.97 d	83.68 d	7.90 i	29.00 a	2.71 a	5.26 e	0.88 ab	6.43 b	68.80 g	6.40 b	51-2	87.52 ^{a-d}
11.	CB-14	28.42 k	23.40 k	82.34 k	9.10 b	28.50 a	2.31 a	5.45 b	0.89 a	6.46 a	65.50 i	6.50 a	61-1	87.42 ^{a-d}
12.	Mutant-1	29.39 g	24.48 g	83.26 g	8.31 f	28.55 a	2.31 a	5.02 g	0.88 ab	5.16 g	70.70 e	6.10 e	51-1	85.49 ^{de}
LSD (5%)		0.007	0.007	0.01	0.005	0.00	0.00	0.007	0.01	0.007	0.007	0.005	-	2.06

Here, the mean values bearing same letter (s) didn't differ significantly at $p < 0.05$, UHML= upper half mean length (mm), ML= mean length (mm), UI= uniformity index (%), SFI= short fiber index, Str.= strength (g/tex), Elong.= elongation (%), Mic.= micronaire (µg/inch), MR= maturity ratio, Moist.= moisture (%), Rd= reflectance, +b= yellowness, GOT= ginning out turn (%) and LSD= Least significant difference at 5% level of significance.

influenced textile grade and consumer preference, confirming the importance of brightness as a selection criterion (Liu *et al.*, 2017; Rauf *et al.*, 2019). Higher yellowness values are generally undesirable as they reduce fabric whiteness and dyeing efficiency (Rauf *et al.*, 2019). These two parameters defined cotton color grade (CG), with the best-performing genotypes (e.g., JA-13/X and BC-0510) showing brighter, less pigmented fiber, corresponding to higher commercial grades (51-1, 51-2).

Ginning out turn (GOT) ranged from 84.98% to 88.90%, with BC-0510 showing the highest performance. Although variation in GOT was relatively narrower compared to fiber quality traits, it remained crucial for economic return, as higher GOT directly increased lint yield (Ali *et al.*, 2019).

Overall, the genotypes BC-0491, BC-0510 and BC-0027 emerged as superior genotypes with consistently high performance across fiber length, uniformity, reflectance, micronaire, maturity and GOT (**Table 27**). Conversely, genotypes such as JA-17/2 and Mutant-1 exhibited lower overall fiber quality and might be excluded from quality-centric breeding pipelines (Patil *et al.*, 2021; Liu *et al.*, 2017). This reinforced the need for trait-specific breeding strategies and multi-location trials to ensure stability in performance. These findings were consistent with earlier reports highlighting the presence of wide genetic variability among upland cotton genotypes and the potential for identifying elite lines for simultaneous improvement of yield and fiber quality (Zhang *et al.*, 2019; Smith & Jones, 2019).

4.3.5 Pearson's correlation matrix among the fiber quality characters

The **Figure 12** presented the Pearson's correlation coefficients among twelve fiber quality parameters of 12 upland cotton genotypes. The analysis revealed distinct patterns of association among fiber length, strength, fineness, and yield-related traits, highlighting both synergistic and antagonistic relationships.

A strong and highly significant positive correlation was observed between upper half mean length (UHML) and mean length (ML) ($r = 1.00$, $p < 0.001$), reflecting their shared contribution to fiber length determination. Similarly, strength (Str) exhibited a near-perfect correlation with elongation (Elong) ($r = 0.99$, $p < 0.001$), suggesting that these two traits were functionally linked and likely controlled by overlapping genetic mechanisms.

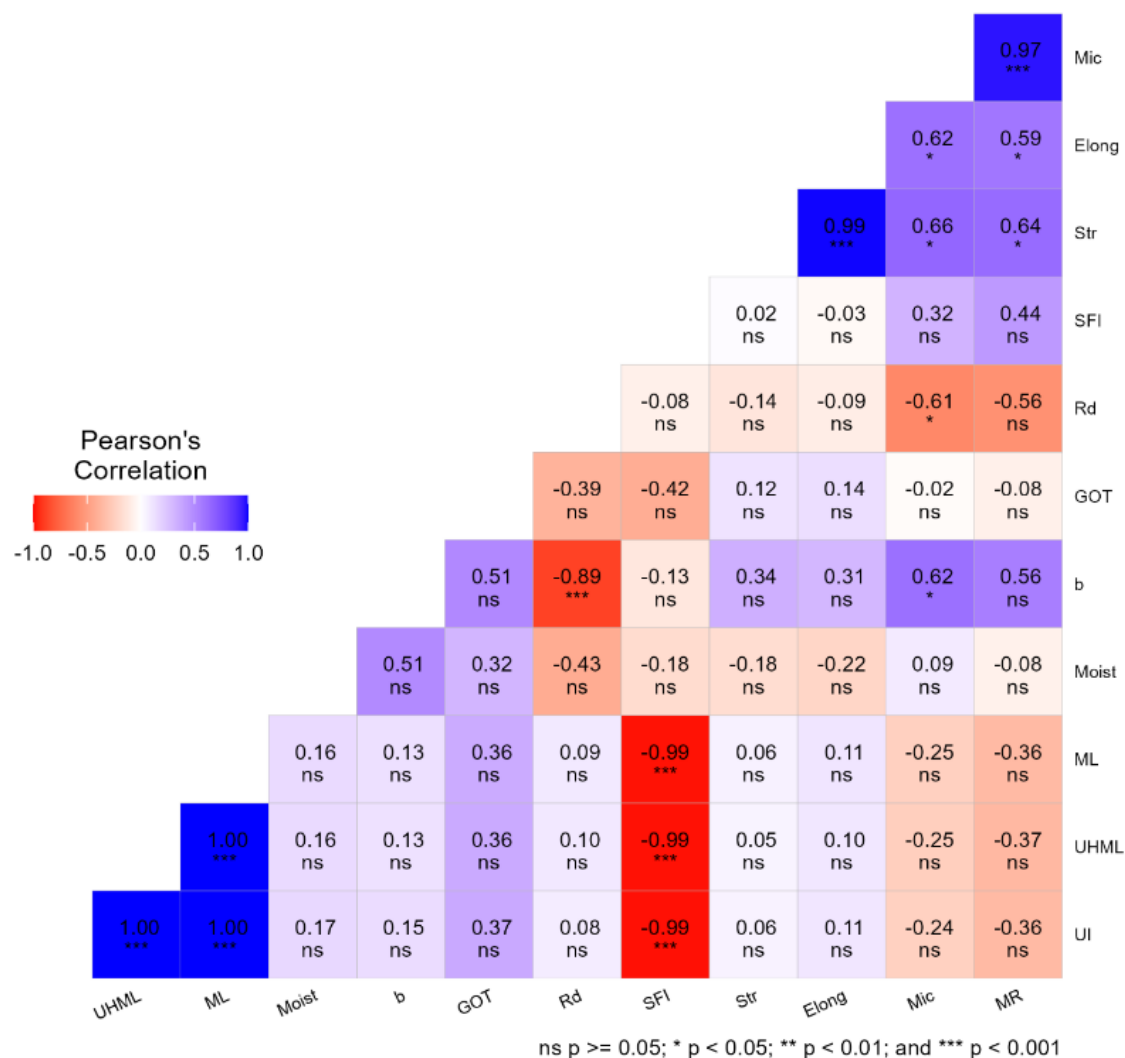


Figure 12. Correlation matrix among the fiber quality parameters in well performing 12 upland cotton genotypes

Here, UHML = upper half mean length (mm), ML = mean length (mm), UI = uniformity index (%), SFI = short fiber index, Str = strength (g/tex), Elong. = elongation (%), Mic. = micronaire ($\mu\text{g}/\text{inch}$), MR = maturity ratio, Moist. = moisture (%), Rd = reflectance, +b = yellowness, and GOT = ginning out turn (%).

Micronaire (Mic) also showed a moderate positive correlation with elongation ($r = 0.62$, $p < 0.05$), aligning with earlier findings that fiber fineness and elongation were often interrelated (Abdurakhmonov *et al.*, 2017; Shang *et al.*, 2016).

In contrast, several negative associations were detected, particularly between yellowness (+b) and reflectance (Rd) ($r = -0.89$, $p < 0.001$), suggesting a trade-off between fiber color parameters. Likewise, short fiber index (SFI) was strongly and negatively correlated with fiber length parameters (ML, UHML, and uniformity index [UI]) ($r = -0.99$, $p < 0.001$) (**Figure 12**), confirming that higher proportions of short fibers adversely affect fiber length and uniformity (Zhang *et al.*, 2019; Sunilkumar *et al.*, 2017). Furthermore, Rd value (fiber reflectance) exhibited a significant negative correlation with micronaire ($r = -0.61$, $p < 0.05$), suggesting that improved fiber reflectance may come at the cost of reduced micronaire value, a trade-off also reported in previous studies (Iqbal *et al.*, 2019). Moisture content (Moist) and maturity ratio (MR) showed weak and non-significant correlations with most parameters, indicating that these traits may be relatively independent of fiber length, strength, or yield traits under the studied conditions.

Overall, the correlation matrix underscored the complexity of cotton fiber quality improvement, where favorable associations (e.g., among length and strength traits) coexisted with trade-offs (e.g., between Rd and GOT, or fiber brightness and strength). Hayat & Bardak opined that correlation analysis showed significant relationship among fiber traits such as fiber length, fiber strength and micronaire in their research. The current results were in agreement with earlier reports emphasizing the need for balanced breeding strategies to optimize multiple fiber quality parameters simultaneously (Fang *et al.*, 2021; Zhang *et al.*, 2019).

4.3.6 Molecular profiling of early boll picking well performing 12 upland cotton genotypes

Forty-five cotton genotypes were grouped into three clusters, later on considering minimization of research waste, improvement of reproducibility and validity of molecular analysis, 12 genotypes were selected representing three clusters. It is noted that careful planning with well-documented protocols might offer more reliable and reproducible results, a fundamental principle for scientific integrity. Hence, molecular

characterization was carried out with well performing 12 upland cotton genotypes possessing earliness.

One of the key motivations for creating the superior upland cotton varieties is the need for increased yield stability, which might be fulfilled by boll setting and maturation being compressed into a shorter duration, more desirable part of the growing season. This section presented the findings from the molecular characterization of well performing twelve upland cotton (*Gossypium hirsutum* L.) genotypes through simple sequence repeats (SSRs) markers, focusing on marker polymorphism, population structure, genetic diversity and similarity, and clustering patterns, with the support of relevant tables and figures.

4.3.7 Polymorphism of SSR Markers

Twenty SSR loci were screened on well performing 12 upland cotton (*Gossypium hirsutum* L.) genotypes. Among them, 9 loci were polymorphic (**Figure 13**) and utilized for subsequent genetic analysis.

The average length of an SSR marker is highly variable in cotton, depending on the concerned locus under interest; however, the length of SSR markers ranged from 100-400 bp but the average amplified length of DNA is more than 800 bp. These molecular tools were applied for genetic profiling to assist further breeding of selected 12 cotton genotypes. The markers constructed genetic maps, detecting quantitative trait loci (QTLs) of each pair of co-dominant alleles, eventually assessed genetic diversity and similarity among 12 cotton genotypes. Such results were furnished as interpreted by Yuzhen *et al.* (2020).

The polymorphic markers generated a total of 26 alleles and an average of 2.89 alleles per locus (**Table 28**). Allele number ranged from one (BNL3479 and FPF1) to five (NAU3736 and MGHE16), and allele sizes ranged between 50 bp and 1100 bp. The polymorphism information content (PIC) values ranged between 0.00 and 0.798 and had an average of 0.501. Among the markers, MGHE16 (0.798), NAU3736 (0.779), and BNL1604 (0.75) were the most informative with high discriminatory power. The estimates indicated a moderate level of polymorphism, which was in agreement with previous findings in upland cotton diversity studies (Shi *et al.*, 2023; Tyagi *et al.*, 2014; Zhang *et al.*, 2013; Fang *et al.*, 2013; Lacape *et al.*, 2007; Zhang *et al.*, 2002; Liu *et al.*, 2000). The moderate mean PIC value (0.501) and presence of several highly informative

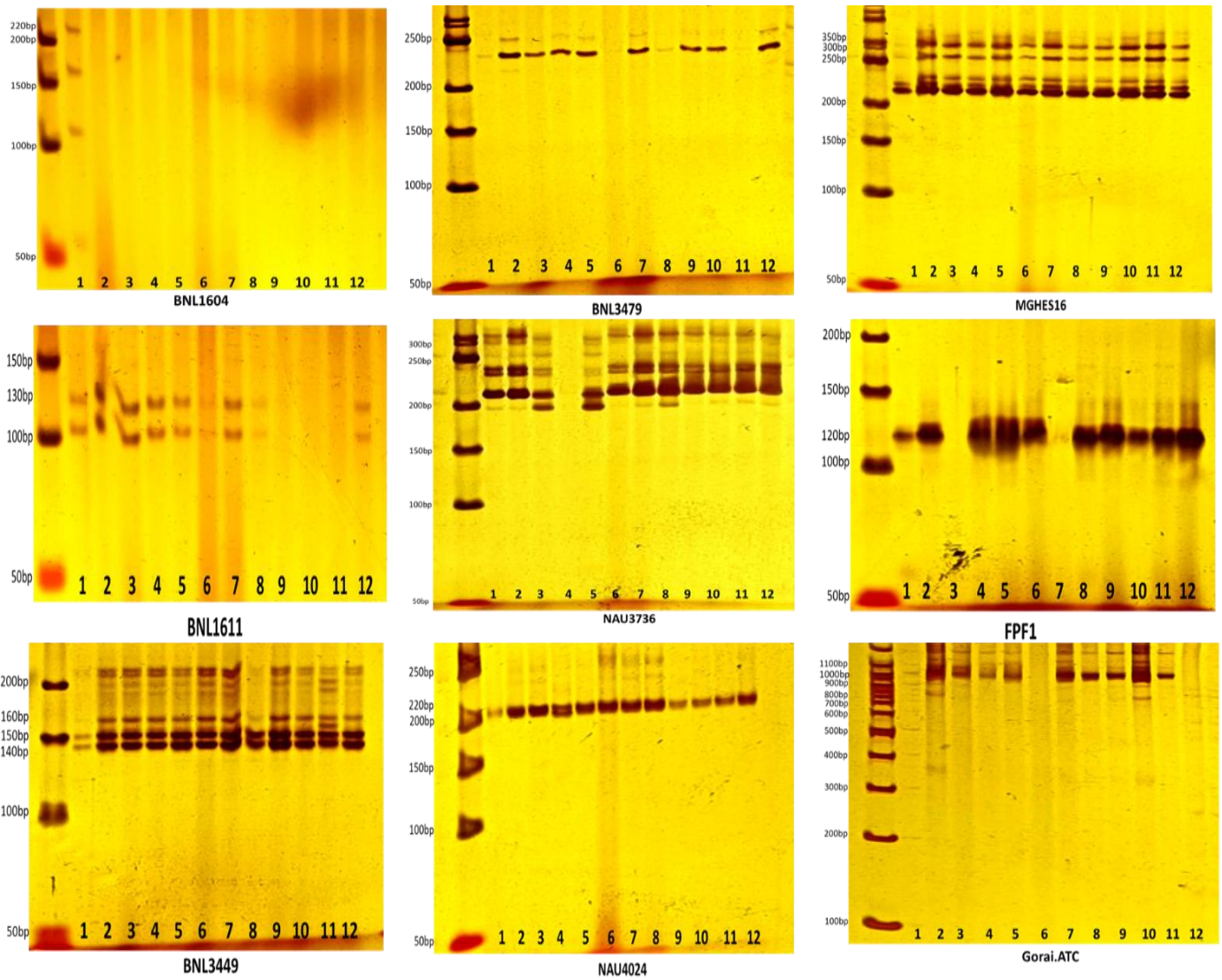


Figure 13. Gel images of the polymorphic microsatellite markers derived from polyacrylamide gel electrophoresis (PAGE) using 50bp and 100bp plus DNA ladder in well performing 12 upland cotton genotypes

Table 28. Number of alleles, allele range and polymorphism information content (PIC values) of the polymorphic markers in well performing 12 upland cotton genotypes

SL.	SSR loci	Number of alleles	Range of allele size (bp)	PIC
1.	BNL1604	4	50-220	0.750
2.	BNL1611	2	100-140	0.500
3.	BNL3449	4	140-220	0.682
4.	BNL3479	1	200-250	0.000
5.	NAU3736	5	200-250	0.779
6.	NAU4024	2	200-220	0.500
7.	MGHES16	5	200-350	0.798
8.	FPF1	1	100-150	0.000
9.	Gorai.ATC	2	900-1100	0.500
Average		2.89		0.501
Total		26		4.509

loci indicated that even a minor collection of SSRs was capable of revealing structure and informing diversity and similarity in well performing 12 cotton genotypes (Lacape *et al.*, 2007; Liu *et al.*, 2000). The polymorphic banding patterns were clearly distinct using PAGE electrophoresis (**Figure 13**), again confirming the efficiency of SSR markers in detecting variability in the genotypes that were tested.

4.3.8 Population structure of early boll picking well performing 12 upland cotton genotypes

Population structure analysis using STRUCTURE software revealed two distinct populations ($K = 2$) as calculated using the ΔK method (Evanno *et al.*, 2005). Population I included genotypes 1, 9, 11, 10 i.e., RA-2, TC-1903, CB-14, and BC-0027 genotypes, while Population II included the other eight genotypes such as 7, 5, 3, 12, 4, 2, 8, 6 viz., JA-17/2, BC-0510, RA-9, N5, Mutant-1, BC-0491, RA-5, JA-13/X and BC-0512 (**Figure 14 & Table 29**). The outcome was presented in **Figure 14**, where the individuals were represented as colored segments i.e., Population I (Red color) and Population II (Green color) according to their population assignment.

The uneven genotypic distribution revealed that Population II was more genetically diverse than the Population I, which was comprising the majority of local checks and almost related lines, has a narrower genetic base. In a similar pattern of clustering, in which the former was broad and diverse while the latter was restricted and homogeneous were also reported in previous molecular characterization studies of cotton (Abdurakhmonov *et al.*, 2008; Mei *et al.*, 2013).

4.3.9 Genetic differentiation and diversity study in early boll picking well performing 12 upland cotton genotypes

Genetic differentiation analysis revealed widespread divergence between the two identified populations. Population II exhibited a high mean F_{ST} of 0.5215, revealing strong differentiation, whereas Population I exhibited negligible differentiation ($F_{ST} = 0.0286$) (**Table 30**). In addition, mean genetic distances between clusters were greater in Population I (0.3582) than in Population II (0.1898). These findings showed that the local checks Mutant-1 and CB-14 were more genetically divergent compared to other lines. This divergence provided an opportunity for the generation of new genetic recombinations through hybridization with Population II genotypes, as suggested by Tyagi *et al.*, 2014; Fang *et al.*, 2013; Lacape *et al.*, 2007.

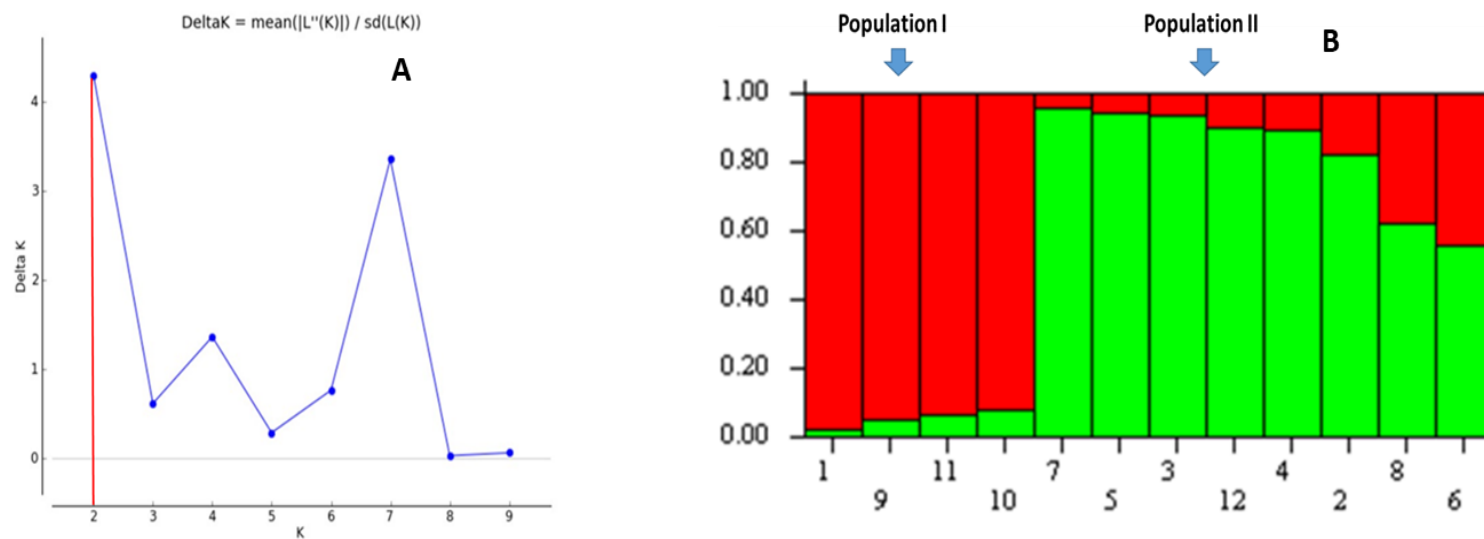


Figure 14. Model based population structure plot depicting A) the optimal number of groups determined by the Evanno methods (Delta K) and B) bar plot from structure cluster analysis with k=2

Here, Color codes: Population I= Red and Population II = Green, 1= RA-2, 2= RA-5, 3= RA-9. N5, 4= BC-0491, 5= BC-0510, 6= BC-0512, 7= JA-17/2, 8= JA-13/X, 9= TC-1903, 10= BC-0027, 11= CB-14 and 12= Mutant-1.

Table 29. The grouping of well performing 12 upland cotton genotypes into two distinct populations

Population	Size	Genotype
I	4	RA-2, TC-1903, CB-14, BC-0027.
II	8	JA-17/2, BC-0510, RA-9.N5, Mutant-1, BC-0491, RA-5, JA-13/X, BC-0512.

Table 30. Mean FST values and average distances between individuals in clusters

Population	Mean values of FST	Average distances between clusters
Population I	0.0286	0.3582
Population II	0.5215	0.1898

4.3.10 Cluster analysis of well performing 12 upland cotton genotypes

UPGMA dendrogram analysis revealed two major clusters (**Figure 15**) reflecting structured genetic dissimilarities. Cluster I included RA-5, RA-9.N5, BC-0491, BC-0510, BC-0512, JA-17/2, JA-13/X, and Mutant-1, shared allelic similarities, suggesting potential redundancy forming a closely related subgroup. Cluster II comprised RA-2, TC-1903, CB-14, and BC-0027, with RA-2 and CB-14 showing higher similarity. The genotypes RA-2, CB-14 in cluster II were more dissimilar, offering complementary alleles for breeding programs. Such consistency among different methods of clustering had also been reported in SSR-based diversity analysis in cotton germplasm (Abdurakhmonov *et al.*, 2017; Peakall & Smouse, 2012; Lacape *et al.*, 2007; Evanno *et al.*, 2005; Pritchard *et al.*, 2000). In Cluster I, BC-0491, BC-0510 and BC-0512 comprised a closely subgroup (Ib), while JA-17/2 and JA-13/X were also part of a close sub-grouping (Ia). Mutant-1 was relatively isolated, but nonetheless, in the same cluster. On the other hand, in Cluster II, RA-2 (IIa) and CB-14 (IIb) shared greater similarity than TC-1903 (IIb) and BC-0027 (IIb) (**Figure 15**). Overall, the early maturing genotypes RA-2, BC-0510, BC-0491, BC-0512 were genotypically different from check varieties CB-14 and Mutant-1 and located in different sub-clusters.

4.3.11 Principal coordinate analysis of early boll picking well performing 12 upland cotton genotypes

Principal coordinate analysis (PCoA) based on PC1 and PC2, revealing clear genetic differentiation and clustering among the genotypes. This pattern reflected structured allelic variation detected by highly polymorphic SSR markers. The majority of the total genetic variation was explained by the first three principal coordinates (**Figure 16**).

In the two-dimensional PCoA plot (**Figure 17A**), the genotypes were clearly separated into two major genetic groups along PC1 and PC2, reflecting pronounced molecular divergence among the evaluated materials. The compact clustering of genotypes within each group suggests higher genetic similarity, whereas the clear inter-group separation indicated substantial allelic differentiation. Such structuring is indicative of distinct genetic backgrounds, likely shaped by differences in parental origin, selection history, or adaptation strategies.

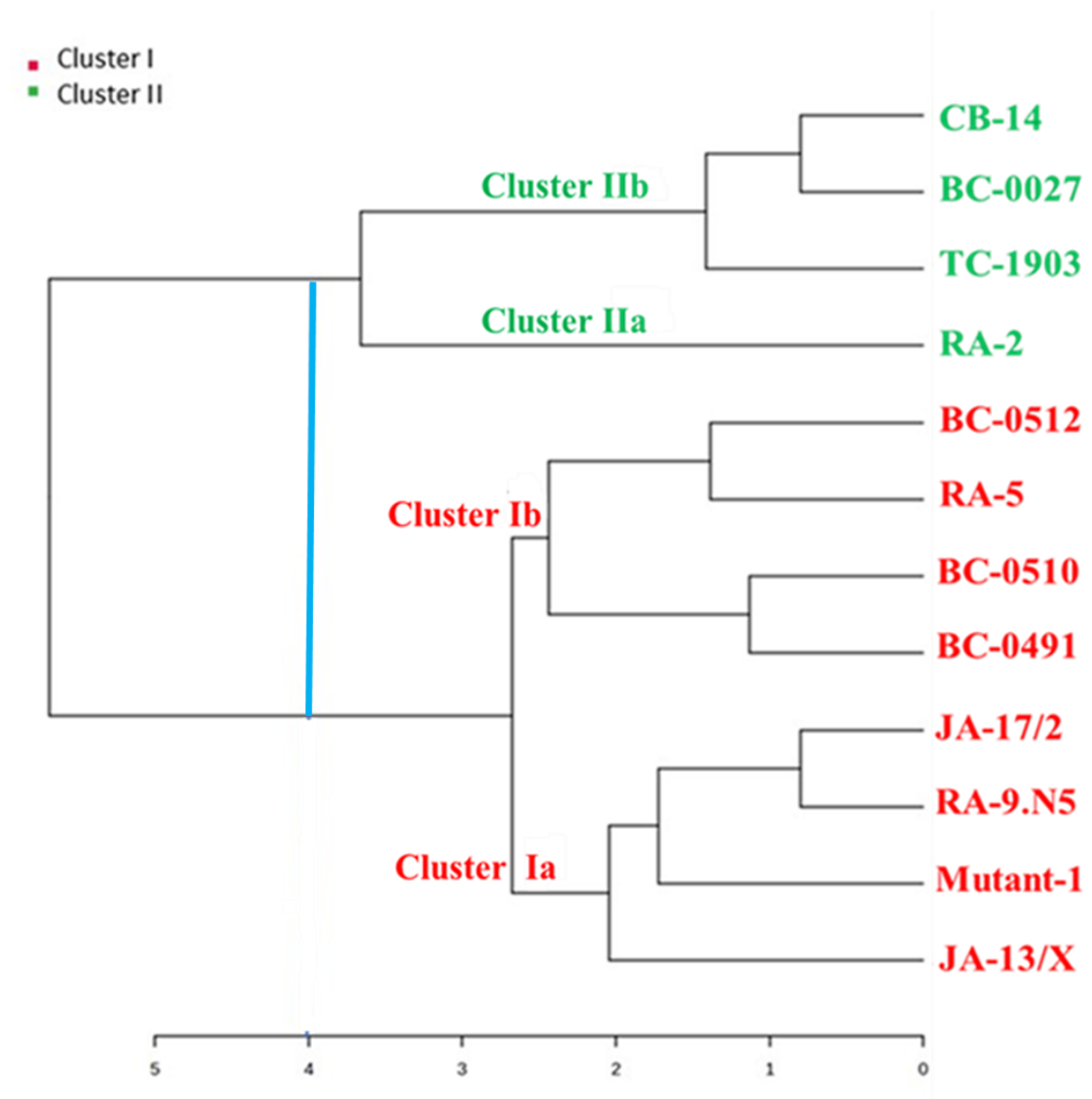


Figure 15. An UPGMA cluster dendrogram showing the genetic relationships in well performing 12 upland cotton genotypes based on alleles detected by 9 microsatellite markers

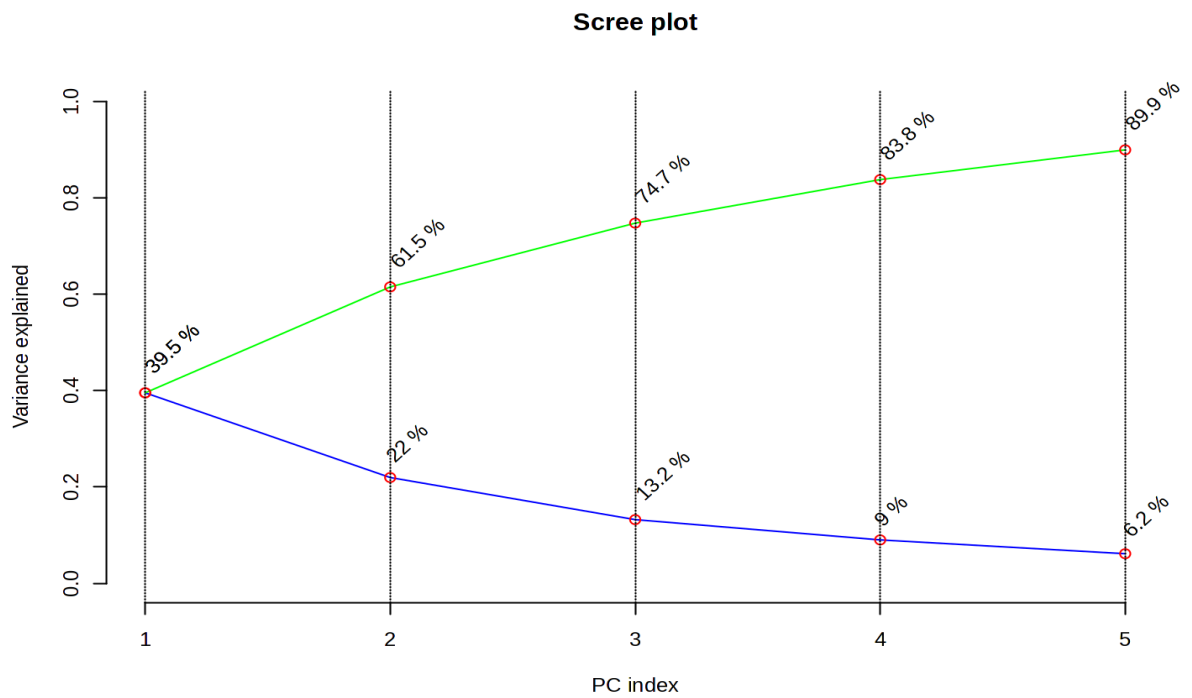


Figure 16. The scree plot displaying principal coordinate analysis of top 5 PCs of well performing 12 upland cotton genotypes based on SSR marker data

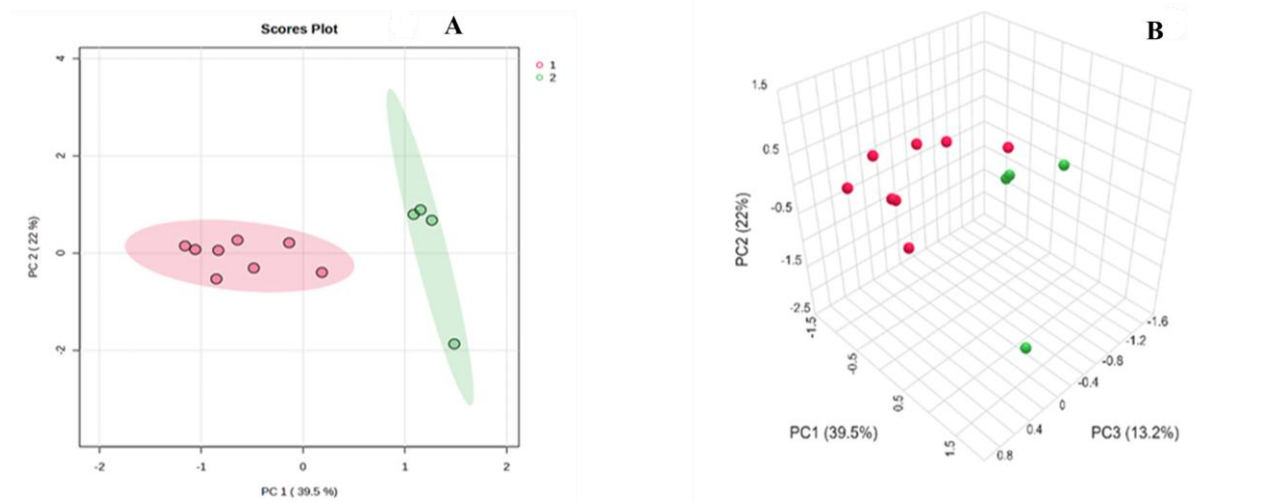


Figure 17. Principal coordinate analysis (PCoA) based on SSR polymorphisms in well performing 12 upland cotton genotypes A) 2-Dimensional and B) 3-Dimensional

The three-dimensional PCoA plot (**Figure 17B**) further resolved the genetic relationships by incorporating PC3, allowing improved visualization of subtle genetic distances that were less apparent in the two-dimensional projection. The spatial dispersion of genotypes across the three axes confirmed the presence of considerable molecular diversity within the panel, supporting the robustness of SSR markers in capturing genome-wide polymorphism in upland cotton.

The PCoA patterns corroborated the clustering results and demonstrated substantial genetic dissimilarities among the well-performing genotypes. This molecular divergence provided valuable opportunities for parental selection in breeding programs aimed at combining early maturity with high yield potential, while simultaneously broadening the genetic base of cultivated upland cotton (Iqbal *et al.*, 2019; Fang *et al.*, 2013; Ulloa M., 2006).

4.3.12 Heatmap visualization of genetic relationships between genotypes and markers

The genic relationships between the well performing twelve upland cotton genotypes were further assessed by a heatmap of nine polymorphic SSR markers. Rows represented the markers, and columns represented the genotypes, thus showing genetic relatedness among genotypes and marker alleles in a single overview (**Figure 18**). The color gradient corresponded to the allelic similarity index, where red indicated higher similarity (positive association) and blue denoted lower similarity or greater genetic distance (negative association).

Two major clusters among the genotypes were identified, in question. A genetically almost alike group was formed by CB-14, BC-0027, TC-1903 and RA-2, whereas a diverged branch was formed by BC-0491, BC-0510, RA-5 and BC-0512 based on their allelic divergence. The allele cluster pointed out other patterns. Certain BNL1604 alleles (i.e., BNL1604c and BNL1604a) correlated strongly with CB-14 within cluster II, whereas NAU3736d and NAU3736e contributed more to TC-1903. On the other hand, different MGHE16 alleles and NAU3736a/b/c contributed more to Mutant-1 and other individuals of cluster I. The Gorai.ATC alleles belonged to the MGHE16 allele and contributed strongest in BC-0491 and Mutant-1, respectively. Similarly, JA-13/X shared a strong association with an allele of BNL3449 and RA-9.N5 was more intense for an allele of BNL1611 (**Figure 18**)

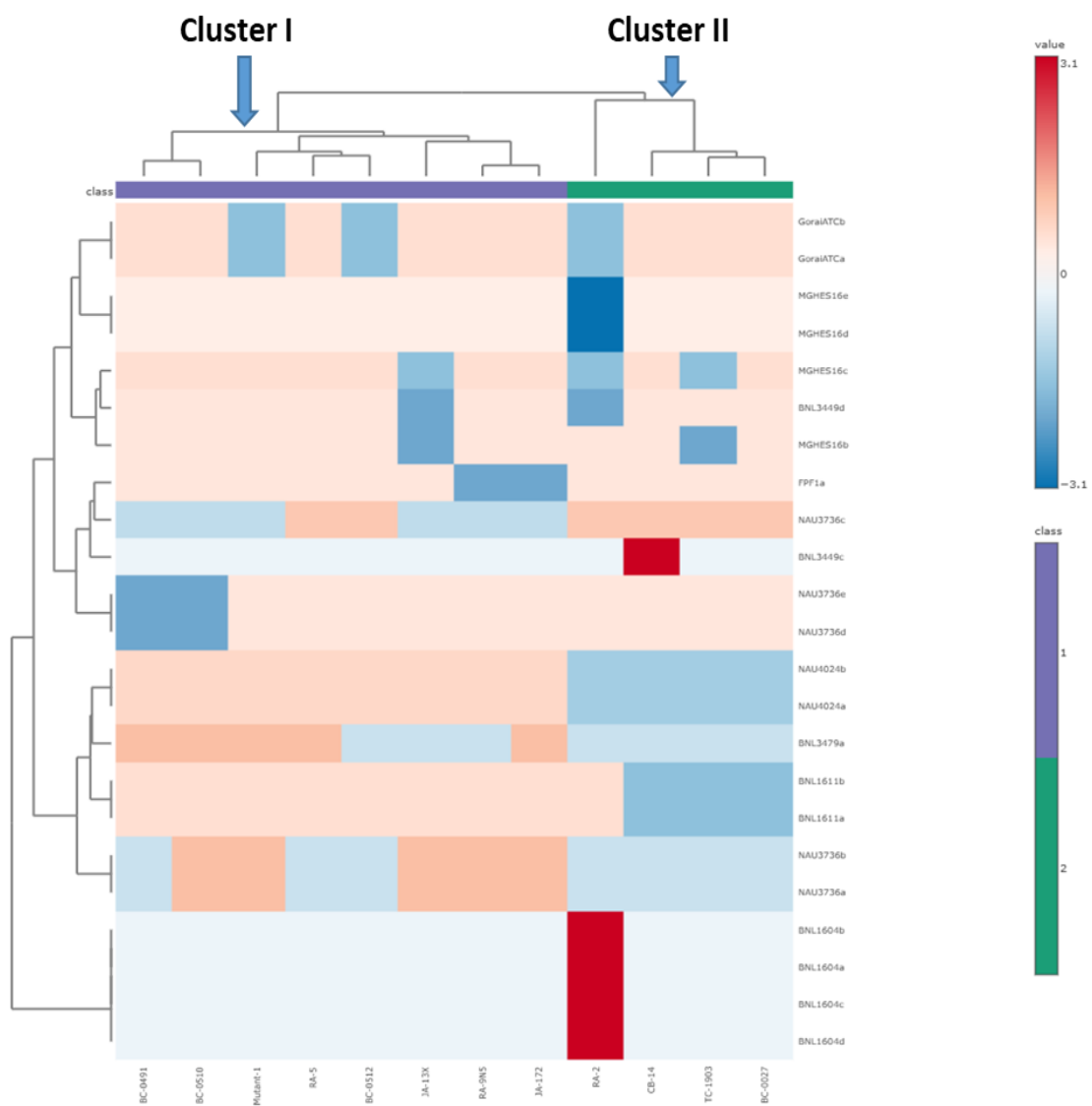


Figure 18. A heat map showing the genetic relationships of well performing 12 upland cotton genotypes and alleles detected by 9 microsatellite markers

Overall, the heatmap facilitated the separation of genotypes into two major clusters and additionally uncovered allele-specific correlations with certain genotypes. Similar cluster genotypes had high similarity values, while distinct lines such as CB-14 and BC-0510 were placed in distant clusters. The results were corroborated by the patterns revealed by dendrogram analyses but additionally provided higher resolution on allele-level differentiation among the genotypes.

These findings demonstrated that SSR markers were effective for assessing genetic relationships in upland cotton. The congruence between PIC profiles, Population STRUCTUREs, UPGMA dendrograms, and PCoA analyses provided a robust molecular framework for selecting genetically divergent parents to maximize heterosis, maintain breeding diversity, and accelerate development of early boll picking, high-yielding cotton cultivars adapted to Bangladesh. This approach aligned with prior recommendations emphasizing molecular tools to complement phenotypic selection for breeding elite cotton genotypes (Abdurakhmonov *et al.*, 2017; Zhang *et al.*, 2013; Abdalla *et al.*, 2001).

4.4 Narrowing down from 100 to 45 to 12 genotypes applying different biometrical tools with proper justification

Most of the data generated in three experiments are field oriented except molecular analysis (**Figure 19**). In the entire investigation the destination is authentication as well as interpretation of data by applying different biometrical tools, like mean performances, post hoc Dunnett's test, genetic parameters, correlation, path analysis, genetic diversity, principal component analysis etc. The main focus of the investigation is identification of early boll picking and high seed cotton yield genotypes. Accordingly, the study was initiated with 100 cotton genotypes and gradually reduced to 45 and then 12 genotypes. In every experiment contributions of seed cotton yield related traits and high fiber potential genotypes are identified. The biplot and the dendrogram highlights significant variation in studied cotton cultivars for different traits and it is assessed that the variation in these genotypes might be exploited to develop early lines along with the increased seed cotton yield having good fiber quality (Sarwar *et al.*, 2020). When the yield enhancing traits are confirmed then molecular tools are applied for valid conclusion. After completion of investigation five outstanding genotypes are achieved which might be further advanced for multi location yield trial with a view to develop high quality and high fiber yielding varieties in near future (**Figure 19**).

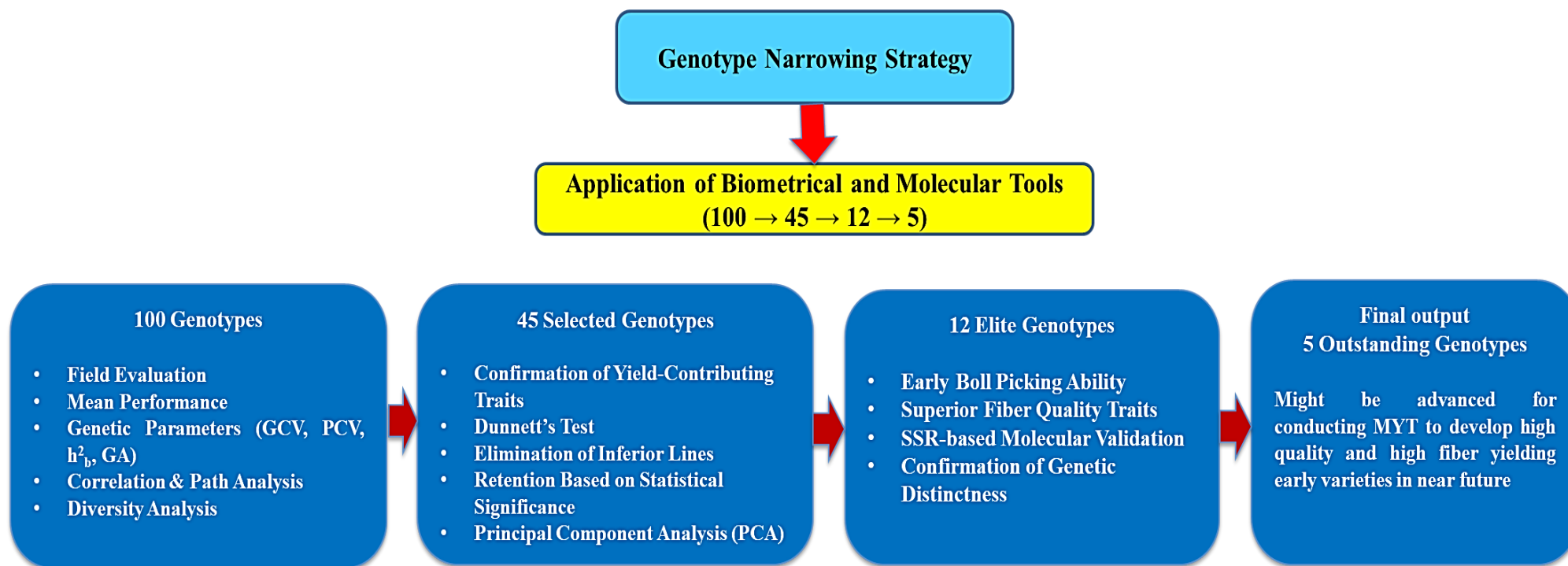


Figure 19. Narrowing down from 100 to 45 to 12 genotypes applying different biometrical tools with proper justification

4.5 Comparison between molecular and phenotypic results for identifying well performing early maturing genotypes

Cluster analysis incorporating both morphological and genetic data revealed similar grouping. High-yielding earliness genotypes (RA-2, RA-5, RA-9.N5, BC-0491, BC-0510, BC-0512, JA-17/2, JA-13/X, TC-1903, BC-0027) clustered together in Cluster I under morphological clustering (**Figure 9**), while all in Cluster I except RA-2, TC-1903, BC-0027 and CB-14 under molecular analysis (**Figure 15**). Conversely, the two check genotypes (CB-14 and Mutant-1) clustered together in Cluster II of the morphological dendrogram (**Figure 9**), but in different clusters in molecular analysis (**Figure 15**). The consistency between molecular and phenotypic clustering confirmed early-maturing, high-yielding genotypes RA-2, BC-0491, BC-0510, BC-0512 was genetically distinct from the check varieties (CB-14, Mutant-1) and located in separate sub-clusters. SSR marker based morphological and molecular outcome also revealed harmony in past studies of cotton (Manivannan *et al.*, 2025). Molecular markers data are efficient for measuring genetic dissimilarities and relatedness and also aid in selection of diverse outstanding lines to be used in future breeding programs to develop new cotton cultivars (Modi *et al.*, 2020).

4.6 Promising lines in upland cotton (*Gossypium hirsutum* L.) from overall studies (2021-23 growing periods)

High seed cotton yield in upland cotton is the ultimate desirable agronomic trait that influenced by several traits such as sympodial branches per plant, bolls per plant, single boll weight, boll bursting percentage, early completion of boll picking etc. If bolls are picked early the crop can avoid the onset of unfavorable climatic conditions, such as terminal drought and of course reduces unburst bolls increase in number due to low temperature for early coming winter, which is a great problem in northern belt of Bangladesh. Early boll picking additionally contributes to efficient resource utilization and facilitates relay cropping patterns.

The present study evaluated earliness with high seed cotton yielding performance of promising cotton lines in comparison with standard check CB-14, using days to 50% flowering (DFF), earliness index (EI %) and other seed cotton yield contributing traits, mentioned earlier. The overall mean results from 2021 to 2023 growing periods are

shown by mean performances of studied traits of 100, 45 and 12 genotypes in 2021, 2022 and 2023, respectively.

Reduction of growth duration at vegetative stage can be achieved by managing sowing time, spacing, fertilizers, irrigation schedule, hormone application etc. and in reproductive stage short duration is achieved mainly by defoliation. These elements are not included in the current study because focused on reduction of duration at reproductive stage considering two major components like DFF and EI (%).

4.6.1 Early performance of the promising lines compared to standard check based on DFF over the experimental tenure (2021-23)

In this context, the **Figure 20** depicted the early performance of the promising lines compared to standard check CB-14 based on variation in days to 50% flowering over the experimental tenure (2021-23). The data are presented as mean $\pm$ standard error (SE), with statistical significance denoted by asterisks ($*p < 0.05$, $**p < 0.01$, $***p < 0.001$).

Genotype CB-14 (Standard check) exhibited the longest flowering duration, requiring approximately 54 days to reach 50% flowering, while RA-2 showed the shortest duration (<48 days) signifying its early-flowering nature, followed closely by BC-0510, BC-0027 and BC-0512, respectively. These genotypes attained 50% flowering earlier than the standard check, demonstrating their potential as early-maturing lines. BC-0491 showed intermediate flowering time, suggesting moderate earliness.

The red bars and asterisks indicated pairwise comparisons demonstrating that CB-14 differed significantly from all other genotypes ($***p < 0.001$ with RA-2 and BC-0027; $**p < 0.01$ with BC-0512; $*p < 0.05$ with BC-0491 and BC-0510). These results suggested substantial genotypic variation in flowering time, which may reflect underlying genetic differences influencing floral transition pathways (Andrés & Coupland, 2012; Bernier & Périlleux, 2005). Early-flowering genotypes like RA-2 possessed allelic variants promoting accelerated floral induction under the tested environmental conditions, a trait often associated with adaptation to short growing seasons or drought-prone environments (Blümel *et al.*, 2015). Overall, the results indicated that CB-14 is a late-flowering genotype, whereas RA-2 represents an early-flowering type, highlighting the potential of genotypic selection for breeding programs aimed at optimizing flowering phenology.

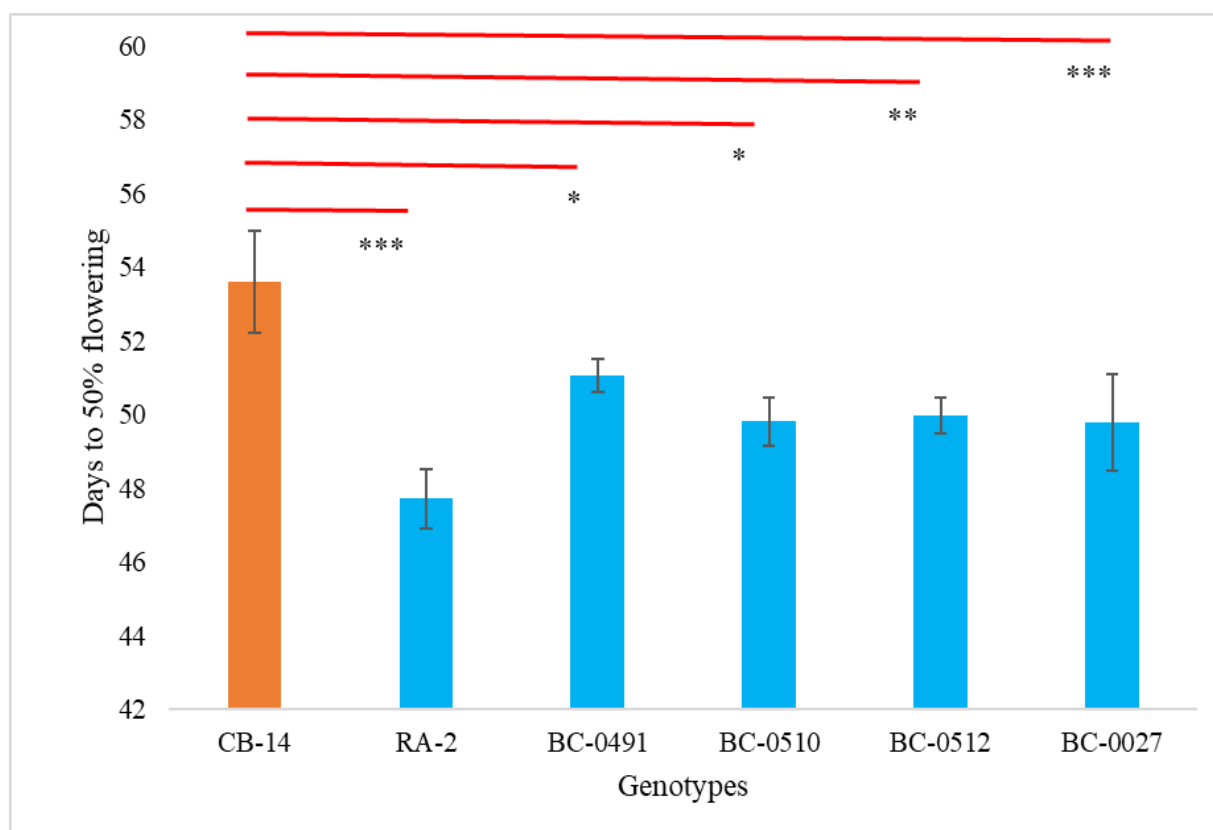


Figure 20. Early performance of the promising lines compared to standard check based on DFF over the experimental tenure (2021-23)

Here, *, **, *** denoted significant at 5%, 1% & 0.1% level of significance, respectively.

Saraswat *et al.*, (2022); Baloch *et al.*, (2014) reported days to first flower expressed changes from 51.6 to 66.6 days and 90 to 120 days, respectively. Bhatti *et al.*, (2020a,b) found that days to 50% flowering varied from 61-70 days, however the lowest and statistically at par value was expressed by the cultivar FH-142 followed by MNH-886. The average days to 50% flowering was reported in other findings as 72.48 days (Raj & Patil, 2023), 60.67 days (Saraswat *et al.*, 2022) and 65.04 days (Sarwar *et al.*, 2021b). Therefore, the selected promising lines revealed early flowering than others that will be helpful to develop earliness varieties in Bangladesh.

4.6.2 Early performance of the promising lines compared to standard check based on EI (%) over the experimental tenure (2021-23)

The **Figure 21** presented the early performance of the promising lines compared to standard check CB-14 based on the variations of earliness index (%) over the experimental tenure (2021-23), expressed as mean $\pm$ standard error (SE). Statistical differences among genotypes are denoted by asterisks (* $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$).

The earliness index serves as a quantitative measure inversely related to the number of days to flowering-higher values indicate earlier flowering and maturity (Baye & Becker, 2005). Genotype RA-2 and BC-0027 exhibited the highest earliness index (>92%), significantly surpassing CB-14 (*** $p < 0.001$) and several other genotypes like BC-0491, BC-0510 and BC-0512 (* $p < 0.05$). CB-14 showed the lowest earliness index (~86%), consistent with its delayed flowering behavior as shown in **Figure 20**. Intermediate values were observed for BC-0491, BC-0510, and BC-0512, suggesting moderate earliness among these lines. The significant pairwise differences highlighted by red bars indicated clear genotypic variation in phenological development.

These results reinforced the contrasting phenological responses observed among the studied genotypes, with RA-2 and BC-0027 classified as early-maturing type and CB-14 as a late-maturing genotype (**Figure 21**). Variation in earliness index likely reflects underlying genetic control of developmental rate, possibly mediated by allelic differences in flowering-time regulatory genes such as *Constans* (CO), *Flowering Locus T* (FT), and *Gigantea* (GI) (Andrés & Coupland, 2012; Blümel *et al.*, 2015). Early-maturing genotypes may possess adaptive advantages under environments prone to terminal drought or heat stress, as shorter phenological cycles enable

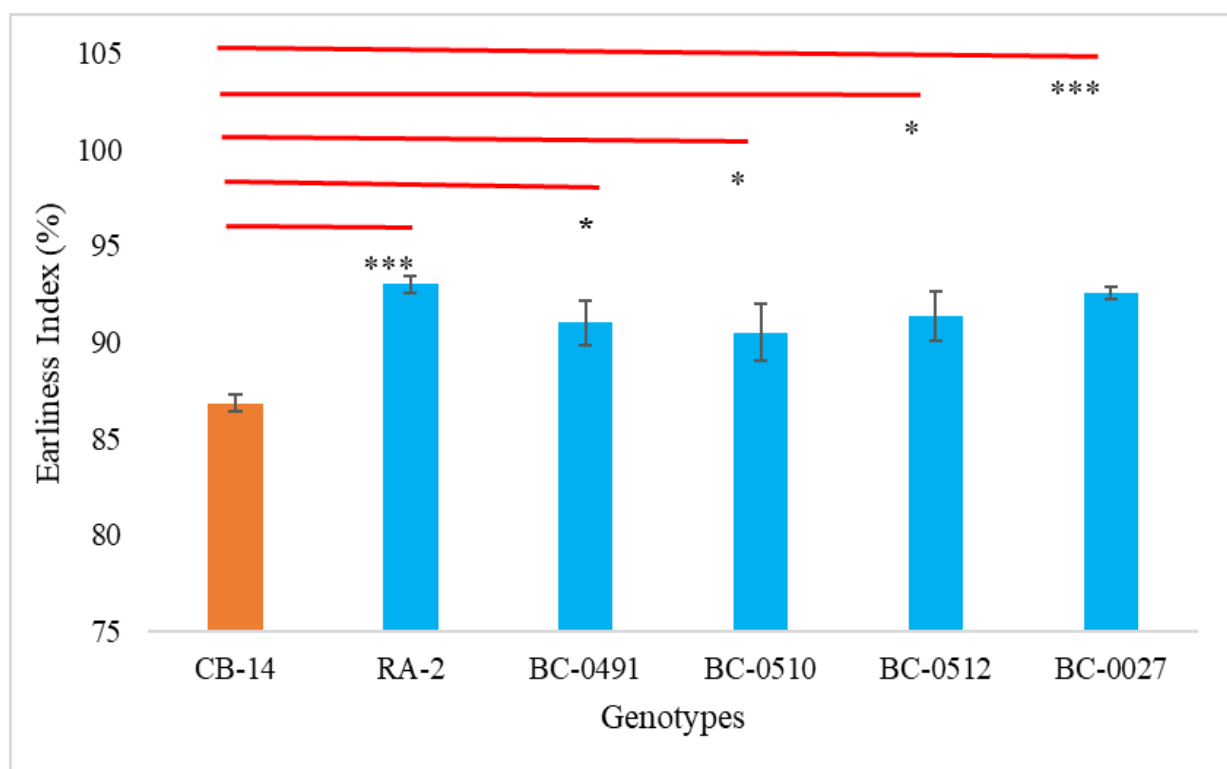


Figure 21. Early performance of the promising lines compared to standard check based on EI (%) over the experimental tenure (2021-23)

Here, *, **, *** denoted significant at 5%, 1% & 0.1% level of significance, respectively.

reproductive success before the onset of adverse climatic conditions (Craufurd & Wheeler, 2009; Kiani *et al.*, 2007; Singh & Kumar, 2020). Amer & Hassan, (2023) found earliness index varied from 58.41% for Giza 87, which indicated more lateness to 82.39% for promising line no.3, the more earliness of this genotype might be attributed to the foreign genotype involved in it. Abubakar *et al.*, (2024) showed that the greater earliness index was recorded in PB-130 (67.06) and the lowest earliness index was recorded in PB-94 (42.73) when analyzed 14 genotypes of cotton. 15.32% earliness percentage had been reported (Raj & Patil, 2023) whereas 57.73% earliness index had also been stated (Mahdi & Emam, 2020) in cotton. Therefore, the selected promising lines revealed higher mean (>90%) of earliness index that will increase the chance of the genotypes to include in earliness cotton variety development.

4.6.3 Yield performance of the promising lines compared to standard check over the experimental tenure (2021-23)

The **Figure 22** illustrated the yield performance of the promising lines compared to standard check CB-14 over the experimental tenure (2021-23) based on mean values of seed cotton yield per plant (g), presented as mean $\pm$ standard error (SE). Significant differences among genotypes are indicated by asterisks (* $p < 0.05$, ** $p < 0.01$).

Substantial genotypic variation was observed in seed cotton yield per plant. CB-14 produced the lowest yield (119.28 g plant⁻¹), while BC-0510 and BC-0512 recorded the highest yields (125.35 g & 124.90 g plant⁻¹). Intermediate yields were obtained from BC-0027, RA-2 and BC-0491, all of which differed significantly from CB-14 (* $p < 0.05$ to ** $p < 0.01$). The significant pairwise differences, as denoted by red bars, underscore the presence of genetic variability influencing yield potential among the evaluated lines.

The relatively low yield of CB-14 corresponds with its delayed flowering and lower earliness index (**Figure 20 & 21**), indicating that prolonged vegetative growth did not translate into higher reproductive output under the given environmental conditions. In contrast, the higher-yielding genotypes, particularly BC-0510 and BC-0512, exhibited both early to moderate flowering and enhanced reproductive efficiency. This suggested that an optimal balance between earliness and effective assimilate partitioning contributes to improved yield performance (Bange *et al.*, 2010;

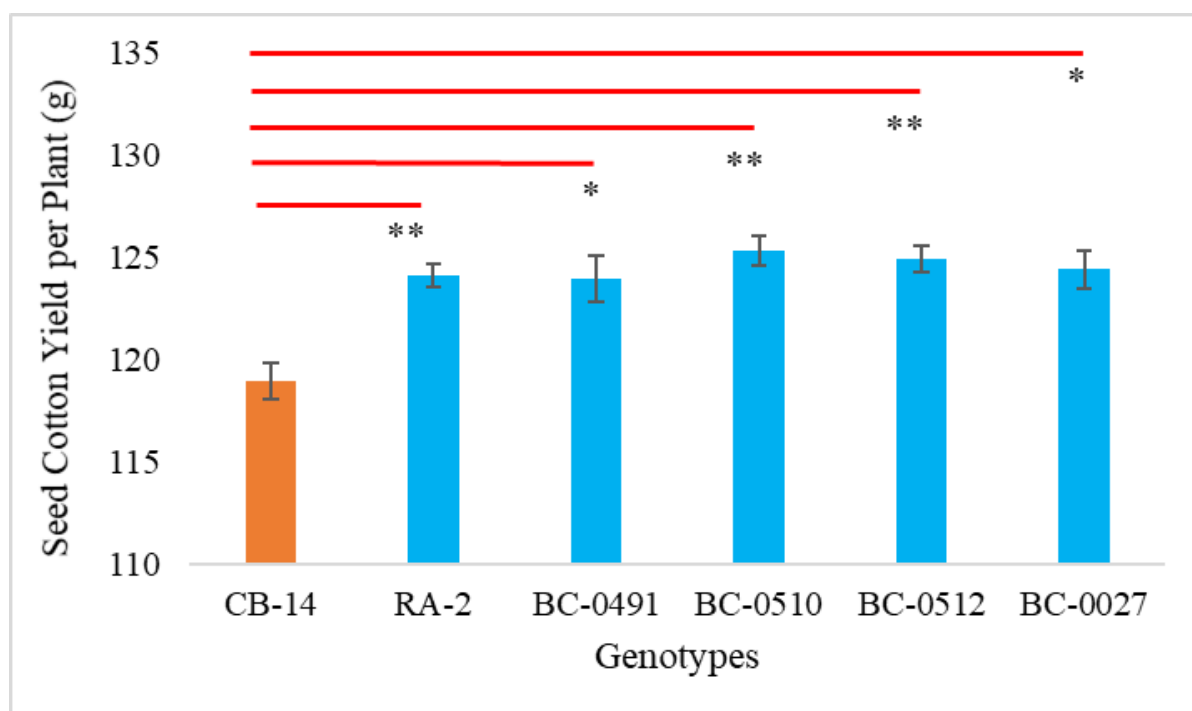


Figure 22. Yield performance of the promising lines compared to standard check over the experimental tenure (2021-23)

Here, *, ** denoted significant at 5% & 1% level of significance, respectively.

Wells & Meredith, 1984). Differences in seed cotton yield may be attributed to genotypic variation in traits such as boll number, boll weight, and photosynthetic capacity during the reproductive phase (Reddy *et al.*, 2017; Pettigrew, 2004). Rehman *et al.*, (2020) examined that IUB-222 scored the highest mean value of 74.81g for seed cotton yield. Mahdy *et al.*, (2023) showed that mean seed cotton yield per plant of the parents Giza90 and Giza86 was 104.67 and 94.06g under normal field condition. Ahsan *et al.* (2015) observed that late-maturing genotypes produced lower yields, reflecting the lower seed cotton yield per plant (SCYPP), lint fiber yield per plant (LFYPP), and earliness index (EI) in CB-14. Overall, these results highlighted significant genetic variability for yield potential among the tested genotypes, with BC-0510 and BC-0512 emerging as promising candidates for yield improvement in breeding programs aimed at enhancing productivity and adaptation.

4.6.4 Ginning out turns (%) of the promising lines compared with two standard checks

Figure 23 presented the ginning out turn (GOT) percentage of the promising cotton genotypes in comparison with two established standard checks, CB-14 and Mutant-1 measured in 2023. Ginning outturn, defined as the percentage of lint obtained from a given weight of seed cotton, is a critical quantitative trait in cotton breeding, directly impacting the economic value of the crop (Smith & Jones, 2019). High GOT is a key objective for breeders due to its strong positive correlation with fiber yield (Ali *et al.*, 2019; Khan *et al.*, 2017).

The data indicated significant phenotypic variability in GOT across the tested genotypes. The checks exhibited moderate GOT values. CB-14 recorded 87.4%, while Mutant-1 showed the lowest value at 85.4%. Whereas, several promising lines demonstrated superior GOT compared to both standard checks. BC-0510 achieved the highest GOT at 88.9%, significantly exceeding both CB-14 (87.4%) and Mutant-1 (85.4%). BC-0512 and RA-2 also showed high performance, recording 88.6% and 87.8%, respectively (**Figure 23**).

The genotypes BC-0510, BC-0512, RA-2, and BC-0027 (87.5%) all surpassed the GOT of the high-performing check, CB-14. This suggested the successful identification of breeding material with enhanced lint productivity (Bardak & Hayat, 2020; Fang *et al.*, 2021; Zhang *et al.*, 2019). Conversely, BC-0491 (87.2%) performed marginally below

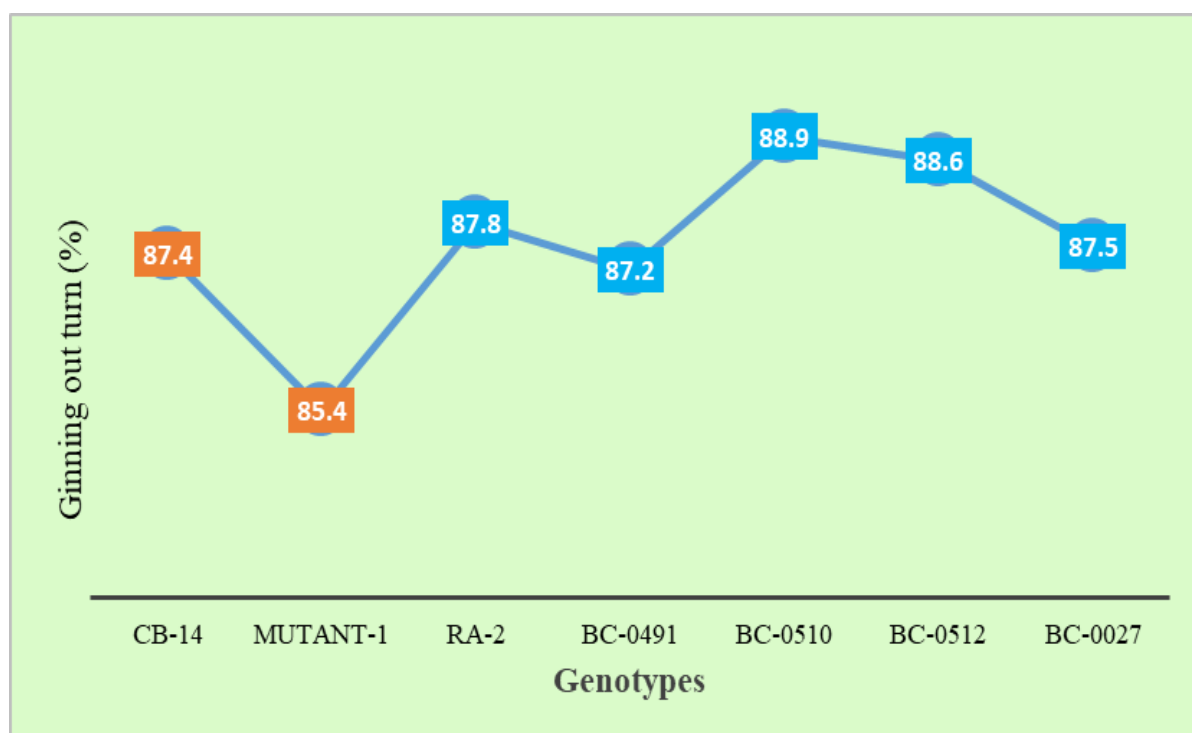


Figure 23. Ginning out turns (%) of the promising lines compared with two standard checks (CB-14 and Mutant-1) measured in 2023

CB-14 but still above Mutant-1. The observed variation in GOT is attributable to underlying genetic control of lint density, fiber length, and seed size, and provides essential data for genotype selection in advanced breeding programs (Patel & Shah, 2018). Genotypes such as BC-0510 and BC-0512 are prime candidates for further evaluation and utilization in crossing programs aimed at developing high-yielding, commercially viable cotton cultivars.

Meanwhile, of the well performing early boll picking 12 genotypes, two population structures were developed through molecular analysis of which genotypes, BC-0510 and BC-0491 under Population II were genetically far away from BC-0027 under Population I. Therefore, simultaneous consideration of DFF, EI, SCYPP, GOT (%) and molecular based population structure, genotypes, RA-2, BC-0491, BC-0510, BC-0512 and BC-0027 might be selected for multilocation yield trial to develop outstanding early cotton varieties.

CHAPTER V

SUMMARY AND CONCLUSION

Upland cotton dominates world production (~90%) due to its broad adaptability, yield potential, and desirable fiber characteristics. Therefore, this research assessed genetic variability, heritability, genetic divergence, trait associations, fiber quality attributes, and molecular characterization among diverse upland cotton genotypes to identify elite parental lines and for developing early maturing high yield potential with superior fiber quality varieties of upland cotton. However, the findings of the entire investigation are described below-

Initially, 100 upland cotton genotypes were evaluated for eleven yield-related traits to estimate the extent of variability, heritability, genetic advance and diversity. The analysis of variance revealed highly significant differences among genotypes for all measured traits, indicating the presence of substantial genetic diversity within the germplasm. Genotypes such as JA-11/L, BC-0025, BC-0033, BC-0027, RA-2, and BC-0024 exhibited superior yield and earliness characteristics. High heritability coupled with high genetic advance for boll number, boll weight, and seed cotton yield suggested the predominance of additive gene action, indicating that these traits could be improved effectively through direct selection. Seed cotton yield per plant (SCYPP) showed significant positive correlations with sympodial branches per plant (SBPP), phenotypic acceptability (PA) and bolls per plant (BPP) at both genotypic and phenotypic levels. Path analysis confirmed that SBPP, BPP, and single boll weight (SBW) exerted strong positive direct effects on yield. These findings emphasize the importance of selecting the right combination of traits to maximize seed cotton yield potential while maintaining fiber quality and earliness.

Cluster analysis classified the 100 genotypes into eight groups, with Cluster VI containing high-yielding genotypes and Cluster II comprising early-maturing lines. Principal component analysis (PCA) revealed that the first five components explained 73.09% of the total variation, and genotypes were broadly scattered in the biplot. The contribution of specific traits to total genetic divergence, as revealed by PCA, indicated that seed cotton yield per plant, bolls per plant, and boll weight were major

discriminating factors, further confirming their significance as key selection criteria in breeding programs.

Further, the evaluation of selected 45 upland cotton genotypes provided important insights into the potential of developing early-boll harvesting cotton varieties suitable for the limited growing season of Bangladesh. The results demonstrated considerable variability for days to flowering and maturity, enabling the identification of genotypes capable of completing their life cycle within a shorter duration without compromising yield potential. Genotypes such as RA-2, BC-0027, BC-0491, BC-0510, and BC-0512 emerged as promising entries, combining earliness with high yield and desirable morphological characteristics. BC-0510, BC-0512, and RA-2 exhibited significantly reduced plant height, higher sympodial branches, earlier flowering, and enhanced boll production. BC-0491 and BC-0027 displayed superior single boll weight and seeds per boll, resulting in higher overall seed cotton and lint fiber yields, while also showing early maturity. Dunnett's test confirmed that RA-5, BC-0419, BC-0510, and BC-0027 significantly outperformed the standard checks (CB-14 and Mutant-1) for several yield and morphological attributes.

Cluster analysis classified the genotypes into three groups, with high-yielding and early-maturing lines such as RA-2, RA-5, BC-0491, BC-0510, BC-0512, and BC-0027 predominantly in Cluster I. These genotypes were identified as potential parents for future breeding due to their balanced performance in yield, minimum days for t boll picking, and boll characteristics. PCA revealed that yield and earliness traits dominated genetic variation. The first three principal components accounted for 87.47% of the total variation, with genotypes scoring high on PC1 and low on PC2 characterized by earliness, high yield, and short stature.

Finally, morphological characterization of the selected well performing 12 upland cotton genotypes highlighted the diversity in plant height, sympodial branches per plant, bolls per plant, single boll weight and early boll maturity. These morphological traits, when considered together with yield-related attributes, provide a useful basis for selecting ideotypes having the characteristics, early boll harvesting, high seed cotton yield with quality fiber suited to specific environments and management practices. The SCYPP trait ranged from 116.35-125.88 g and the highest SCYPP was recorded from BC-0510 followed by RA-2 with 125.24g. Overall earliness indices were appreciable among 12

genotypes, ranged from 87.65-93.27%. On the basis of such performances, selection of 12 genotypes for quality characters study and molecular analysis was an authentic decision.

The analysis of fiber quality traits of the selected 12 lines showed that significant genotypic variation exists for properties such as upper half mean length (UHML), fiber strength, micronaire, and reflectance. Genotypes BC-0491 and BC-0510 consistently exhibited superior fiber characteristics such as high UHML, strength, and reflectance, making them potential candidates for use as donor parents in fiber quality improvement programs. The positive correlations among fiber length, strength, and reflectance indicated that simultaneous improvement of these traits is feasible through selection, thereby ensuring that yield gains are not achieved at the expense of fiber quality. The integration of fiber traits into yield-based selection strategies will be essential to produce balanced cultivars meeting both agronomic and industrial requirements. Collectively, BC-0491, BC-0510, JA-17/2, BC-0512, RA-2, and BC-0027 represent valuable genetic resources for enhancing fiber quality in breeding programs.

Molecular profiling using simple sequence repeat (SSR) markers provided a complementary perspective on the genetic dissimilarities identified through phenotypic evaluations. Out of 20 SSR markers, nine were polymorphic, producing 26 alleles. Markers MGHES16 and NAU3736 generated the maximum number of alleles (five each) and exhibited the highest polymorphic information content (PIC = 0.798 and 0.779, respectively), reflecting substantial genetic dissimilarities. Population STRUCTURE revealed two distinct genetic groups (K=2) among twelve genotypes. Population II exhibited higher dissimilarities, while Population I was more homogeneous. UPGMA dendrogram analysis also identified two major groups, wherein early-maturing, high-yielding genotypes (RA-2, BC-0510, BC-0491, BC-0512) were genetically distinct from the check varieties (CB-14, Mutant-1) and located in separate sub-clusters. Crosses between divergent populations are suggested to maximize heterosis and broaden the genetic base. Heatmap visualization further illustrated genetic relationships at the allele level, revealing allele-specific associations with certain genotypes and confirming the presence of two major groups. These analyses demonstrated clear separation between the more homogeneous Population I and the diverse Population II. The findings underscore the usefulness of molecular markers in

validating phenotypic diversity and in guiding the selection of genetically dissimilar parents for hybrid development.

Across all experiments, results demonstrate significant genetic variability among upland cotton genotypes for yield, earliness, and fiber quality traits. The combination of morphological, phenotypic, and molecular analyses provided a holistic understanding of the genetic resources available for cotton improvement in Bangladesh. High heritability and genetic advance for yield traits confirmed additive gene effects, indicating that conventional selection could be effective. Elite genotypes such as JA-11/L, RA-2, BC-0027, BC-0491, BC-0510, and BC-0512 consistently exhibited superior performance across multiple experiments. Integration of multivariate and molecular analyses validated the phenotypic diversity observed and identified genetically dissimilar yet complementary parental lines for hybridization and selection programs.

This research provides a robust genetic, phenotypic, and molecular foundation for the improvement of upland cotton in Bangladesh. The identified superior genotypes, trait associations, and molecular diversity patterns offer clear guidance for designing targeted breeding strategies aimed at developing resilient, high-yielding, early-boll picking, and high-quality cotton cultivars suited to local agro-ecological conditions. The integrated approaches combining field evaluation, statistical analysis, and molecular tools can effectively guide breeding decisions and accelerate varietal development. Ultimately, the outcomes of this research lay the groundwork for a more resilient and productive cotton sector in Bangladesh, capable of meeting both national and global demands for high-quality fiber.

RECOMMENDATIONS

The findings of this study provide a valuable genetic foundation for breeding programs aimed at developing early-boll picking, high fiber yielding, and superior fiber-quality producing cultivars. Therefore, five outstanding genotypes viz. RA-2, BC-0491, BC-0510, BC-0512 and BC-0027 might be adopted for MYT to develop early boll picking, high seed cotton yielding and having high GOT (%) to meet up the demand for desirable early varieties of cotton growers of our country.

REFERENCES

- Aarathi V, Balu PA, Premalatha N. (2018) Genetic variability studies in germplasm of upland cotton (*Gossypium hirsutum* L.). *Electron J Plant Breed.* **9**(4):1600-5.
- Abbas HG, Mahmood A, Ali Q. (2015) Genetic variability and correlation analysis for various yield traits of cotton (*Gossypium hirsutum* L.). *J Agric Res.* **53**(4):481-90.
- Abdalla A, Reddy O, El-Zik K, *et al.* (2001) Genetic diversity and relationships of diploid and tetraploid cottons revealed using AFLP. *Theor Appl Genet.* **102**(2–3):222–9.
- Abdel-Monaem MA, Abido WA, Hadházy Á, *et al.* (2022) Genetic divergence among Egyptian cotton genotypes under water deficit conditions. *Acta Ecol Sinica.* **42**(2):11-8.
- Abdurakhmonov IY, Devor EJ, Buriev ZT, Huang L, Makamov A, Shermatov SE, *et al.* (2017) Fiber quality and productivity traits in *Gossypium hirsutum* L.: QTL mapping and candidate gene identification. *BMC Genomics.* **18**(1):1–18.
- Abdurakhmonov IY, Kohel RJ, Yu JZ, Pepper AE, Abdullaev AA, Kushanov FN, *et al.* (2008) Molecular diversity and association mapping of fiber quality traits in exotic *G. hirsutum* L. germplasm. *Genomics.* **92**(6):478–87.
- Abro TF, Kaloi AA, Sootaher JK, Shar PA, Baloch TA, Soomro TA, *et al.* (2021) Assessment of genetic variability in F₄ populations of upland cotton (*Gossypium hirsutum* L.). *Sarhad J Agric.* **37**(3):722–34.
- Abubakar M, Zahid M, Raheem F, Ain NU, Rafi A, Akram Z, *et al.* (2024) Genetic Variability for Earliness Traits in Upland Cotton. *Trends Biotechnol Plant Sci.* 115-22.
- Ahmad S, Ahmad S, Ashraf M, Khan NU, Iqbal N. (2008) Assessment of yield-related morphological measures for earliness in upland cotton (*Gossypium hirsutum* L.). *Pak J Bot.* **40**(3):1201–7.
- Ahmad S, Fiaz S, Riaz A, Bashir I, Zeb A. (2016) Correlation analysis of morphological and fiber quality traits in upland cotton (*Gossypium hirsutum* L.). *Int J Biosci.* **9**(4):200-8.

- Ahmed W, Khan NU, Khalil MR, Parveen A, Saeed M, Shah SA. (2008) Genetic variability and correlation analysis in upland cotton. *Sarhad J Agric*. **24**(4):573-80.
- Ahmed MM, Guo H, Huang C, Zhang X, Lin Z. (2013) Selection of core SSR markers for fingerprinting upland cotton cultivars and hybrids. *Aust J Crop Sci*. **7**(12):1912-20.
- Ahmed S. (1995) *Muslin: Our Glorious Heritage*. Dhaka: University Press Limited.
- Ahsan MZ, Laghari MA, Bhutto H, Soomro AW, Majidano MS, Malik HT. (2015) Genetic variability in different biometric traits of upland cotton genotypes. *Pak J Agric Agric Eng Vet Sci*. **31**(2):215–25.
- Akter T, Islam AK, Rasul MG, *et al.* (2019) Evaluation of genetic diversity in short duration cotton (*Gossypium hirsutum* L.). *J Cotton Res*. **2**:1-6.
- Alam SN, Sarker D, Pradhan MZH, Rashid MH, Sarkar MA, Begum K, *et al.* (2018) First report of occurrence of fall armyworm *Spodoptera frugiperda* in Bangladesh. *Bangladesh J Entomol*. **28**:97-101.
- Alehegn A, Fentahun M, Tsegaye Y. (2020) Performance evaluation of cotton (*Gossypium hirsutum* L.) varieties for yield and related traits in North-Western Ethiopia. *East Afr Scholars J Biosci Biol*. **1**(1):1–5.
- Ali MA, Khan IA, Awan SI, Shiraz A, Shahid N. (2008) Genetics of fibre quality traits in cotton (*Gossypium hirsutum* L.). *Aust J Crop Sci*. **2**(1):10-17.
- Ali MMA, Islam MK, Khalequzzaman MA, Uddin MF. (2019) Evaluation and Characterization of Upland Cotton Genotypes. *Int J Sustain Agril Tech*. **15**(7):01-5.
- Ali S, Ijaz A, Zafar A, Ashraf A, Yunus M, Iqbal Z, Nadeem S, Rajoka MI. (2011) Investigation of simple sequence repeats (SSR) marker-assisted genetic diversity among upland Bt- and non Bt-cotton varieties. *Afr J Biotechnol*. **10**:15222-8.
- Al-Jibouri H, Miller PA, Robinson HF. (1958) Genotypic and environmental variances and covariances in an upland Cotton cross of interspecific origin 1. *Agron J*. **50**(10):633-6.
- Allard RW. (1960) *Principles of plant breeding*. New York, USA: John Wiley and Sons Incorporation.
- Altaf MT, Jamil A, Iqbal J, Latif MS, Nuruzzaman M, Fahad M, Zayed EM. (2024) Exploring the genetic diversity of upland cotton (*Gossypium hirsutum* L.) genotypes using EST-SSR markers. *Indian J Exp Biol*. **62**(12):1010-1070.

- Amer EA, Hassan SS, Abo-Hegazy SRE. (2021) Genetic analysis for yield and fiber quality traits in Egyptian cotton (*Gossypium barbadense* L.). *Egypt J Plant Breed.* **25**(2):177–201.
- Amer EA, Hassan SS. (2023) Genetic Variability among Egyptian Extra-Long Staple Cotton Genotypes for Some Economic Traits. *Egypt J Agron.* **45**(2):189-200.
- Amer EA. (2022) Identifying Superior Parents and Hybrids for Yield, Its Components and Fiber Quality in Cotton (*Gossypium barbadense* L.). *Egypt J Agron.* **44**(1):59-73.
- Amin MR. (2018) Rejuvenation, Characterization and Evaluation of Cotton Germplasm. CRC Rangpur. Annual Research Report Breeding 2009-2010.
- Andrés F, Coupland G. (2012) The genetic basis of flowering responses to seasonal cues. *Nature Reviews Genet.* **13**(9), 627–639.
- Araújo WP, Pereira JR, Zonta JH, Guerra HOC, Cordão MA, de Lima RF. (2019) Fiber quality in upland cotton cultivars under water deficit strategies. *Afr J Agric Res.* **14**(23):999-1011.
- Asha R, Ahamed ML, Babu DR, Kumar PA. (2013) Multivariate analysis in upland cotton (*Gossypium hirsutum* L.). *Madras Agric J.* **100**(4-6):333-335.
- Aydin A, Gucer EU. (2023) Genetic Diversity of Some Upland and Sea Island Cotton Genotypes by High Resolution Capillary Electrophoresis Gel. *Preprints.* 5-11.
- Baghyalakshmi K, Priyanka RA, Sarathapriya G, Ramchander S, Prakash AH. (2024) Genetic improvement of fiber quality in tetraploid cotton: an overview of major QTLs and genes involved in and edited for the quality of cotton fibers. *J Cotton Res.* **7**(1):33.
- Balarabe A, Yahaya AI, Mohammed SB, Mohammed MS. (2022) Genetic diversity for earliness, fibre quality and yield components in cotton (*Gossypium spp*). *Biol Environ Sci J Trop.* **19**(3):1-10.
- Balçı Ş, Çınar VM, Ünay A. (2020) A Study on genetic advance and heritability for quantitative traits in cotton (*Gossypium hirsutum* L.). *Adnan Menderes Üniversitesi Ziraat Fakültesi Dergisi.* **17**(1):81-4.
- Baloch MJ, Kumar C, Jatoi WA, Rind IH. (2014) Phenotypic correlation and regression analysis of yield and fibre traits in upland cotton (*Gossypium hirsutum* L.). *Pak J Agric Agric Eng Vet Sci.* **30**(2):135-46.
- Baloch M, Baloch AW, Baloch MK, Mallano IA, Baloch AM, Baloch NJ, Abro S. (2015) Association and heritability analysis for yield and fiber traits in promising

- genotypes of cotton (*Gossypium hirsutum* L.). *Univ Res Jour. (Sci. Ser.)* **47**(2):303-306.
- Bange MP, Caton SJ, Milroy SP. (2010) Managing partitioning and growth in cotton crops to maximize yield and quality. *Field Crops Res.* **119**(1):1–9.
- Bano M, Shakeel A, Khalid MN, Ahmad NH, Sharif MS, Kanwal S, *et al.* (2023) Estimation of Combining Ability for Within-Boll Yield Components in Upland Cotton (*Gossypium hirsutum*). *Sarhad J Agric.* **39**(1).
- Batool S, Khan NU, Makhdoom K, Bibi Z, Hassan G, Marwat KB, *et al.* (2010) Heritability and genetic potential of upland cotton genotypes for morpho-yield traits. *Pak J Bot.* **42**(2):1057-64.
- Baye T, Becker HC. (2005). Genetic variability and interrelationship of traits in the Ethiopian mustard (*Brassica carinata* A. Braun). *Genetic Resources and Crop Evol.* **52**(4):551–558.
- Beegum S, Hassan MA, Reddy KN, Reddy V, Reddy KR. (2025) Assessing fiber quality variability among modern upland cotton cultivars and incorporating it into the GOSSYM-based fiber quality simulation model. *J Cotton Res.* **8**(1):18.
- Bernier G, Périlleux C. (2005). A physiological overview of the genetics of flowering time control. *Plant Biotechnol J.* **3**(1):3–16.
- Bhailume MS, Borole DN, Magar NM. (2016) Correlation and path analysis between seed cotton yield and its attributing characters studies in deshi cotton. *J Cotton Res Dev.* **30**(1) 29-31.
- Bhatti MH, Yousaf MI, Ghani A, Arshad M, Shehzad A, Mumtaz A, *et al.* (2020a) Assessment of genetic variability and traits association in upland cotton (*Gossypium hirsutum* L.). *Int J Bot Stud.* **5**(2):148-51.
- Bhatti MH, Yousaf MI, Munir M, *et al.* (2020b) Genetic variation and association among upland cotton genotypes under semi-arid conditions. *Int J Biol Biotechnol.* **17**(4):693-699.
- Bilal N, Tipu AL, Liaqat S, Sarwar M, Basheer M. (2024b) Diversity and correlation studies of intraspecific hybrids for their morphological traits in cotton (*Gossypium hirsutum* L.). *Res Square.* 1-24.
- Bilal N, Nazeer W, Tipu AL, Liaqat S, Sarwar M, Ali A, Hussain K, Basheer M. (2024a) Evaluation of Cotton Advanced Lines/Varieties for Genetic Diversity and Correlation Studies of Cotton Leaf Curl Virus Disease with Yield Contributing Traits in Cotton. *J Bioresour Manag.* **11**(3):8.

- Biz BD Review. (2025) Bangladesh to remain world's top cotton importer in MY26-
USDA. <https://bizbdreview.com/bangladesh-to-remain-worlds-top-cotton-importer-in-my26-usda>.
- Blümel M, Dally N, Jung C. (2015) Flowering time regulation in crops- what did we learn from *Arabidopsis*? *Curr Opin Biotechnol.* 32, 121–129.
- Bradow JM, Davidonis GH. (2000) Quantitation of fiber quality and the cotton production-processing interface: a physiologist's perspective. *J Cotton Sci.* 4(1):34-64.
- Brown-Guedira GL, Thompson JA, Nelson RL, *et al.* (2000) Evaluation of genetic diversity of soybean introductions and North American ancestors using RAPD and SSR markers. *Crop Sci.* 40(3):815-23.
- Brubaker CL, Bourland FM, Wendel JF. (1999) The origin and domestication of cotton. In: Smith CW, Cothren JT, editors. Cotton: Origin, History, Technology, and Production.
- Burton GW, De Vane EH. (1953) Estimating heritability in tall Fescue (*Festuca arundinacea*) from replicated clonal material. *Agron J.* 45:478-81.
- Burton GW. (1952) Quantitative inheritance in grasses.
- Çakmak F, Çınar VM, Balci Ş, *et al.* (2023) Yield and fiber quality balance in upland cotton (*Gossypium hirsutum* L.) breeding. *Bd J Bot.* 52(2):283-90.
- Çakmak F, Çınar VM, Balcı Ş, Ünay A. (2024) The Performance of Multi-Parental Cotton (*Gossypium hirsutum* L.) Hybrid Genotypes. *Kahramanmaraş Sütçü İmam Üniversitesi Tarım ve Doğa Dergisi.* 27(4):901-9.
- Castetter EF, Underhill RM. (1935) The Ethnobiology of the Zuni Indians. Albuquerque: University of New Mexico Press.
- Çelik S. (2022) Genetic Diversity Analysis of Some Upland Cotton (*Gossypium hirsutum* L.) Genotypes Using SSR Markers. *Türk Doğa ve Fen Dergisi.* 11(1):80-9.
- Chapara R, Reddy KS, Rani MS, Sudhamani K, Grace AD, Lakshmi NV, *et al.* (2024) Studies on Genetic Divergence through Principal Component Analysis and Mahalanobis D² Analysis for Seed Cotton Yield and Important Fiber Traits in *G. hirsutum* L. *Bt Cotton. Res Square.* (Preprint) 1-22.
- Chapepa B, Mubvekeri W, Mare M, Kutwayo D. (2020) Correlation and path coefficient analysis of polygenic traits of upland cotton genotypes grown in Zimbabwe. *Cogent Food Agric.* 6(1):1823594.

- Chaudhari M, Faldu G, Ramani H. (2017) Genetic variability, correlation and path coefficient analysis in cotton (*Gossypium hirsutum* L.). *Adv Biomed Res.* **8**(6):226-33.
- Chen ZJ, Scheffler BE, Dennis E, *et al.* (2007) Toward sequencing cotton (*Gossypium*) genomes. *Plant Physiol.* **145**(4):1303-10.
- Chong J, Xia J. (2018) MetaboAnalystR: A R package for flexible and reproducible analysis of metabolomics data. *Bioinform.* **34**(24):4313-4.
- Chunthaburee S, Dongsansuk A, Sanitchon J, *et al.* (2016) Physiological and biochemical parameters for evaluation and clustering of rice cultivars differing in salt tolerance at seedling stage. *Saudi J Biol Sci.* **23**(4):467-77.
- Comstock RE, Robinson HF. (1952) Genetic parameters, their estimation and significance. *Proc 6th Int Grassland Congr.* **1**:284-91.
- Cotton Development Board (CDB). (2007) Strategic Plan for Cotton Production in Bangladesh. Dhaka: CDB.
- Cotton.org. (2025) Cotton: From Field to Fabric- Ginning. <https://www.cotton.org/pubs/cottoncounts/fieldtofabric/gin.cfm>
- Cotton Marker Database (CMD), <http://www.cottonmarker.org>
- Craufurd PQ, Wheeler TR. (2009) Climate change and the flowering time of annual crops. *J Exp Bot.* **60**(9):2529–2539.
- Dabholkar AR. (1992) Statistical tools for agricultural research. New Delhi: Ashok Kumar and Sons.
- Dahiphale KD, Deshmukh JD, Bagade AB, Jadhav AB. (2015) Studies on genetic variability, correlation and path coefficient analysis in cotton (*Gossypium hirsutum* L.). *Int J Trop Agric.* **33**(1):23-9.
- De Mendiburu F, Simon R. (2015) Agricolae-Ten years of an open source statistical tool for experiments in breeding, agriculture and biology. *PeerJ PrePrints.* **3**:e1404v1. Preprint.
- De Montellano BO. (2012) Plant Evidence for Contact Between Africa and the New World. *Hall Maat.* 1-19.
- Desai A, Chee PW, Rong J-K, May OL, Paterson AH. (2006) Chromosome structural changes in diploid and tetraploid A genomes of *Gossypium*. *Genome*, **49**..
- Deshmukh MR, Deosarkar DB, Chinchane VN, Deshmukh SB. (2019) Studies of genetic parameters for yield, yield contributing and fiber quality characters in desi cotton (*Gossypium arboreum* L.). *J Cotton Res Dev.* **33**(1):44-9.

- Dewey DR, Lu K. (1959) A correlation and path-coefficient analysis of components of crested wheat grass seed production. *Agron J.* **51**(9):515-8.
- Dhamayanathi KPM, Manickam S, Rathinavel K. (2010) Genetic variability studies in *Gossypium barbadense* L. genotypes for seed cotton yield and its yield components. *Electron J Plant Breed.* **1**(4):961-5.
- Dhivya R, Amalabalu P, Pushpa R, Kavithamani D. (2014) Variability, heritability and genetic advance in upland cotton (*Gossypium hirsutum* L.). *Afr J Plant Sci.* **8**(1):1-5.
- Dinakaran E, Thirumeni S, Paramasivam K. (2012) Yield and fibre quality components analysis in upland cotton (*Gossypium hirsutum* L.) under salinity. *Ann Biol Res.* **3**(8):3910-5.
- Divya M, Sabitha N, Vishnuvardhan KM. (2023) Studies on genetic variability for yield and fibre quality traits in upland cotton (*Gossypium hirsutum* L.). *J Res ANGRAU.* **51**(3):59-64.
- Dristy SA, Dhar AR, Uddin MT. (2024) Sustainable practices for cotton production in Bangladesh: economic and environmental perspectives. *Discov Agric.* **2**:53.
- Earl DA, VonHoldt BM. (2012) STRUCTURE HARVESTER: a website and program for visualizing STRUCTURE output and implementing the Evanno method. *Conserv Genet Resour.* **4**:359-61.
- Erande CS, Kalpande HV, Deosarkar DB, Chavan SK, Patil VS, Deshmukh JD, *et al.* (2014) Genetic variability, correlation and path analysis among different traits in desi cotton (*Gossypium arboreum* L.). *Afr J Plant Sci.* **9**(29):2278-86.
- Eswari KB, Sudheer K, Gopinath S, Rao MVB. (2017) Genetic variability heritability and genetic advance studies in cotton. *Int J Dev Res.* **7**(1):10902-4.
- Evanno G, Regnaut S, Goudet J. (2005) Detecting the number of clusters of individuals using the software STRUCTURE: A simulation study. *Mol Ecol.* **14**(8):2611–20.
- Falconer DS, Mackay FCT. (1996) Introduction to Quantitative Genetics. 4th ed. England: Longman Group Ltd. p. 122-5.
- Fang D, Hinze L, Percy R, Deng D, Thyssen G. (2013) A microsatellite-based genome-wide analysis of genetic diversity and linkage disequilibrium in Upland cotton (*Gossypium hirsutum* L.) cultivars from major cotton-growing countries. *Euphytica.* **191**(3):391–401.

- Fang L, Wang Q, Hu Y, Jia Y, Chen J, Liu B, *et al.* (2021) Genomic analyses in cotton identify signatures of selection and loci associated with fiber quality and yield traits. *Nat Genet.* **53**(3):345–54.
- FAO. (2023) FAOSTAT: Crops and Livestock Products. Rome: Food and Agriculture Organization of the United Nations.
- Farias FJC, de Carvalho LP, da Silva Filho JL, Teodoro PE. (2016) Correlations and path analysis among agronomic and technological traits of upland cotton. *Genet Mol Res.* **15**(3).
- Farooq J, Anwar M, Rizwan M, Riaz M, Mahmood K, Mahpara S. (2015) Estimation of correlation and path analysis of various yield and related parameters in cotton (*Gossypium hirsutum* L.). *Cotton Genomics Genet.* **6**.
- Farooq J, Rizwan M, Sharif I, *et al.* (2017) Genetic diversity studies in some advanced lines of *Gossypium hirsutum* L. for yield and quality related attributes using cluster and principle component analysis. *AA Bioflux.* **9**(3):111-8.
- Fryxell PA. (1979) The Natural History of the Cotton Tribe (*Malvaceae*, Tribe *Gossypieae*). College Station: Texas A&M University Press.
- Fu XQ, Yang FX, Lu XK, Wang XG, Yang BX, Liu FJ, *et al.* (2017) Molecular identification of variety purity in a cotton hybrid with unknown parentage using DNA-SSR markers. *Genet Mol Res Gmr.* **16**:1-8.
- Gibely RHAO. (2021) Estimating genetic variance components and identification of transgressive segregants for two intraspecific extra-long staple cotton crosses. *Menoufia J Plant Prod.* **6**:53-70.
- Glade EH Jr, Collins KJ, Rogers CD. (1981) Cotton Quality Evaluation: Testing Methods and Use. Washington, D.C.: U.S. Department of Agriculture, ERS-668.
- Gnanasekaran M, Thiyagu K, Gunasekaran M. (2018) Genetic variability heritability and genetic advance studies in cotton (*Gossypium hirsutum* L.). *Electron J Plant Breed.* **9**(1):377-82.
- Goldwasser Y, Miryamchik H, Sibony M, Rubin B. (2012) Detection of resistant chickpea (*Cicer arietinum*) genotypes to *Cuscuta campestris* (field dodder). *Weed Res.* **52**(2):122–30.
- Gowda SA, Fang H, Tyagi P, Bourland F, Dever J, Campbell BT, *et al.* (2024) Genome-wide association study of fiber quality traits in US upland cotton (*Gossypium hirsutum* L.). *Theo Appl Genet.* **137**(9):214.

- Grover CE, Gallagher JP, Wendel JF. (2012) Phylogenetic insights into the origin of allotetraploid cottons. *Mol Phylogenet Evol.* **63**:369–81.
- Gulhane A, Wadikar MS. (2017) Correlation analysis of cotton derived through introgressive hybridization. *Bioinfolet.* **14**(1):64-6.
- Hawkins JS, Kim H, Nason JD, Wing RA, Wendel JF. (2006) Differential lineage specific amplification of transposable elements is responsible for genome size variation in *Gossypium*. *Genome Res*, **16**:1252-1261.
- Hayat K, Bardak A. (2020) Genetic variability for ginning outturn and association among fiber quality traits in an upland cotton global germplasm collection. *Sains Malaysiana.* **49**(1):11-8.
- Hendrix B, Stewart JM. (2005) Estimation of the nuclear DNA content of gossypium species. *Ann Bot (Lond)*, **95**:789-797.
- Hinze LL, Horn PJ, Kothari N, Dever JK, Frelichowski J, Chapman KD, *et al.* (2015) Nondestructive measurements of cottonseed nutritional trait diversity in the US National Cotton Germplasm Collection. *Crop Sci.* **55**:770–82.
- Hu Y, Zhang J, Wang Y, Zhu X, Li Y. (2022) Genomic analyses reveal the genetic basis of early maturity and identification of loci and candidate genes in upland cotton (*Gossypium hirsutum* L.). *New Phytol.* **236**(5):1888–903.
- Huang C, Nie X, Shen C, You C, Li W, Zhao W, *et al.* (2017) Population structure and genetic basis of the agronomic traits of upland cotton in China revealed by a genome-wide association study using high-density SNPs. *Plant Biotechnol J.* **15**(11):1374-86.
- Huang G, Wu Z, Percy RG, Bai M, Li Y, Frelichowski JE, *et al.* (2020) Genome sequence of *Gossypium herbaceum* and genome updates of *Gossypium arboreum* and *Gossypium hirsutum* provide insights into cotton A-genome evolution. *Nat Genet.* **52**, 516–524.
- Hussein EH, Osman MH, Hussein MH, Adawy SS. (2007) Molecular characterization of cotton genotypes using PCR-based markers. *J Appl Sci Res.* **10**:1156-69.
- ICAC. (2022) Cotton Data Book 2022. Washington, D.C.: International Cotton Advisory Committee.
- ICAC's (International Cotton Advisory Committee) 82nd Plenary Meeting. (2024) World Cotton Trade 2024.

- Ijaz A, Ali S, Ijaz U, Smiullah, Shaheen T. (2013) Molecular characterization of cotton using simple sequence repeat (SSR) markers and application of genetic analysis. *Int J Genet Mol Biol.* **5**(4):49-53.
- Iqbal J, Altaf MT, Jan MF, Raza W, Liaqat W, ul Haq I, *et al.* (2023) Exploring genetic diversity in cotton genotypes using EST-SSR and ISSR markers: A comparative study. *Sarhad J Agric.* **39**(4):800-14.
- Iqbal M, Khan N, Khan TM, Saba N. (2019) Genetic analysis of fiber quality traits in upland cotton under diverse environments. *Euphytica.* **215**(7):1–14.
- IRRI (International Rice Research Institute) (2014). *Standard Evaluation System for Rice (SES)* (5th ed.). Los Baños, Laguna, Philippines: IRRI.
- Isong A, Balu PA, Ramakrishnan P. (2017) Association and principal component analysis of yield and its components in cultivated cotton. *Electron J Plant Breed.* **8**(3):857-64.
- Jamil M, Ahmad S, Ullah HMZ, *et al.* (2022) Diversity analysis in upland cotton genotypes for fibre quality, disease reaction and yield-related traits. *Plant Cell Biotechnol Mol Biol.* **23**(25-26):59-66.
- Jangid K, Sangwan O, Sagar, Chaudhary M. (2022) Correlation and path analysis for seed cotton yield and its contributing traits under irrigated conditions of Sirsa in desi cotton. *J Agri Search.* **9**(2):129-132.
- Jarwar AH, Iqbal MS, Wang X, Wang L. (2019a) Assessment of genetic variability and association in yield and quality parameters of upland cotton (*Gossypium hirsutum* L.). *Int J Biosci.* **15**(1):462-74.
- Jarwar AH, Wang X, Iqbal MS, *et al.* (2019b) Genetic divergence on the basis of principal component, correlation and cluster analysis of yield and quality traits in cotton cultivars. *Pak J Bot.* **51**(3):1143-8.
- Jarwar AH, Wang X, Wang L, Ma Q, Fan S. (2018) Genetic advancement, variability and heritability in upland cotton (*Gossypium hirsutum* L.). *J Environ Agric Sci.* **16**:24-31.
- Jaslam PKM, Bhardwaj N, Devi M, Singh VK, Anu. (2022) Diversity structure analysis based on hierarchical clustering method. AIP Conference Proceedings 2357, 080003. <https://doi.org/10.1063/5.0080621>
- Jogender OS, Kumar D, Shreya AD. (2023) Assessment of genetic variability in promising Asiatic cotton (*Gossypium arboreum* L.) genotypes under rainfed conditions. *Pharma Innov J.* **12**(2):1642-6.

- Johnson HW, Robinson HF, Comstock RE. (1955) Estimates of genetic and environmental variability in soybean. *Agron J.* **47**:314-8.
- Kadam K, Chinchane VN, Rani SD. (2024) Correlation and path analysis of yield and fiber quality traits in upland cotton (*Gossypium hirsutum* L.). *Int J Adv Biochem Res.* **8**(3):61-6.
- Kakar MA, Baloch AW, Sootaher JK, *et al.* (2021) Assessment of genetic divergence and character association in upland cotton (*Gossypium hirsutum* L.) genotypes. *Int J Biol Biotechnol.* **18**(2):321-7.
- Kalivas A, Xanthopoulos F, Kehagia O, Tsaftaris AS. (2011) Agronomic characterization, genetic diversity and association analysis of cotton cultivars using simple sequence repeat molecular markers. *Genet Mol Res.* **10**:208-17.
- Kalpande HV, Bhale SD, Kale UV, Deshmukh JD, Gite VK, Kakde SS. (2008) Genetic variability and correlation studies in F3 generation of cotton (*Gossypium hirsutum* L.). *Int J Plant Sci.* **3**:94-7.
- Kanta CM, Chamundeswari N, Reddy K, Sreekanth B. (2025) Study of Correlation and Path Coefficient Analysis for Seed Cotton Yield, Yield Attributing and Fibre Quality Traits in Upland Cotton (*Gossypium hirsutum* L.). *J Adv Biol Biotechnol.* **28**(10):515-26.
- Kartesz JT. (1994) A synonymized checklist of the vascular flora of the United States, Canada, and Greenland. 2nd ed. Portland, OR: Timber Press.
- Kassambara A, Mundt F. (2017) Package ‘factoextra’. Extract and visualize the results of multivariate data analyses. **76**(2).
- Khakwani K, Asif M, Ahmed Z, Sarwar G, Abbassi Y, Murtaza G. (2022) Yield and Fiber Quality Traits Study in Upland Cotton (*Gossypium hirsutum* L.) Using Line× Tester Analysis. *Pak J Agric Res.* **35**(3).
- Khan A, Ullah I, Ahmad S, Khan MA. (2020) Genetic diversity for fiber quality traits in upland cotton (*Gossypium hirsutum* L.). *J Plant Breed Genet.* **8**(2):25–32.
- Khan IA, Khan NM, Ullah MA, Gul S. (2003) Genetic variability and correlation analysis in upland cotton. *J Agric Biol Sci.* **22**(1):669–76.
- Khan MA, Ahmed N, Ali M. (2019) Impact of boll bursting on cotton yield and quality. *J Cotton Res.* **42**(3):215–23.
- Khan MI, Haq HA, Ullah K, Arshad M, Majid A. (2017) Genetic Diversity and Correlation Studies for Cotton Leaf Curl Disease (CLCuD), Fiber & Yield

- Related Attributes in Exotic Lines of *Gossypium arboreum* L. *Am J Plant Sci.* **8**(3):615-24.
- Khan N, Khan MF, Iqbal J. (2015) Genetic variability in different biometric traits of upland cotton genotypes. *Int J Biol Biotechnol.* **12**(4):575–80.
- Khan NU, Marwat KB, Hassan G, Farhatullah SB, Makhdoom K, Ahmad W, *et al.* (2010) Genetic variation and heritability for cotton seed, fiber and oil traits in *Gossypium hirsutum* L. *Pak J Bot.* **42**(1):615-25.
- Kiani M, Abasi S, Mohammad A. (2007) Correlation and path coefficient analysis for earliness and yield traits in cotton (*Gossypium hirsutum* L.). *Am J Plant Sci.* **2**(4):341–4.
- Killi F, Beycioglu T. (2020) Yield and Yield Parameters of 46 Cotton (*Gossypium spp.*) Cultivars under Kahramanmaras (Turkey) Conditions. *Int J Environ Agric Res.* **6**(8):13-8.
- Kranthi KR. (2018) Cotton production practices: snippets from global data 2017. *The ICAC Recorder.* **36**, 4-14.
- Kuang M, Yang WH, Xu HX, Wang YQ, Zhou DY, Feng XA. (2011) Construction of DNA fingerprinting and analysis of genetic diversity with SSR markers for cotton major cultivars in China. *Sci Agric Sin.* **44**:20–7.
- Kuang Z, Xiao C, Ilyas MK, Ibrar D, Khan S, Guo L, *et al.* (2022) Use of SSR markers for the exploration of genetic diversity and DNA finger-printing in early-maturing upland cotton (*Gossypium hirsutum* L.) for future breeding program. *Agron J.* **12**(7):1513.
- Kumar CPS, Prasad V, Joshi JL, Rajan RE, Thirugnanakumar S. (2019) Studies on genetic variability, heritability and genetic advance in cotton (*Gossypium hirsutum* L.). *Plant Arch.* **19**(1):934-7.
- Kumar KB. (2020) Inter Relationship between Seed Cotton Yield and Yield Contributing Characters in American Cotton (*Gossypium hirsutum* L.). *Int J Curr Microbiol App Sci.* **9**(6):2276-9.
- Kumari VN, Gunasekaran P. (2019) Evaluation of Genetic Diversity in Cotton (*Gossypium barbadense* L.) Germplasm for Yield and Fibre Attributes by Principle Component Analysis. *Int J Curr Microbiol App Sci.* **8**(04):2614-21.
- Lacape JM, Dessauw D, Rajab M, Noyer JL, Hau B. (2007) Microsatellite diversity in tetraploid *Gossypium* germplasm: assembling a highly informative genotyping set of cotton SSRs. *Mol Breed.* **19**(1):45–58.

- Latif A, Bilal M, Hussain SB, Ahmad F. (2015) Estimation of genetic divergence, association, direct and indirect effects of yield with other attributes in cotton (*Gossypium hirsutum* L.) using biplot correlation and path coefficient analysis. *Trop Plant Res.* **2**(2):120-6.
- Li C, Chen B, Xu X, Li D, Dong J. (2018) Simple sequence repeat markers associated/linked with agronomic traits, as core primers, are eminently suitable for DNA fingerprinting in Upland cotton. *Breed Sci.* **68**(4):393-403.
- Li CQ, Li HP, Zhao HH, Dong N, Wang QL, Ai NJ. (2014) SSR core primers reveal DNA fingerprinting of 30 Upland cotton (*Gossypium hirsutum* L.) varieties in China. *Acta Bot Bor-Occid Sin.* **34**:2210–9.
- Li F, Fan G, Lu C, Xiao G, Zou C, Kohel RJ, Ma Z, Shang H, Ma X, Wu J, *et al.* (2015) Genome sequence of cultivated Upland cotton (*Gossypium hirsutum* TM-1) provides insights into genome evolution. *Nat Biotechnol.* **33**:524–530.
- Liang C, Sun B, Meng Z, Meng Z, Wang Y, Sun G, *et al.* (2017) Co-expression of GR79 EPSPS and GAT yields herbicide-resistant cotton with low glyphosate residues. *Plant Biotechnol J.* **15**(12):1622-9.
- Liang Z, Lv YD, Cai CP, Tong XC, Chen XD, *et al.* (2012) Toward allotetraploid cotton genome assembly: integration of a high-density molecular genetic linkage map with DNA sequence information. *BMC Genomics.* **13**:539.
- Liaquat S, ahmed R, Rafiq M, Ahmad S, Qayyum A, rafiq M, *et al.* (2015) Correlation and path analysis for genetic divergence of morphological and fiber traits in upland cotton (*Gossypium hirsutum* L.). *Int J Agron.* 86-94.
- Liu J, Yu J, Wang C, Zhu H. (2017) Genetic basis of cotton fiber quality and its improvement: A review. *Euphytica.* **213**:231.
- Liu S, Cantrell RG, McCarty JC Jr, Stewart JM. (2000) Simple sequence repeat-based assessment of genetic diversity in cotton race stock accessions. *Crop Sci.* **40**(5):1459–69.
- Local media/trade press. (2025) Bangladesh's cotton consumption in MY 2024-25 is forecasted to rise to 7.8 million bales, with ~7.7 million of those expected to be imported. textiletradebuddy.com+2smartinfoindia.com+2
- Mahdi AH, Emam SM. (2020) Correlation and path coefficient analysis of some earliness measures in Egyptian cotton. *J Plant Prod.* **11**(5):407-11.

- Mahdy E, Ahmed A, Abdel-Zaher G, Sayed M, Husein M. (2018) Genetic analysis of earliness and lint yield under normal and late sowing dates in Egyptian cotton. *Egypt J Agron.* **40**(1):31–44.
- Mahdy EE, Mahrous H, Sayed MA, Housein MG. (2023) Observed Genetic Gain from Selection in Egyptian Cotton (*G. barbadense* L) Under Normal and Saline Soils. *Egypt J Agron.* **44**(2):191-208.
- Makhmadjanov S, Tokhetova L, Daurenbek N, Dyamurshayeva G, Makhmadjanov D. (2024) Selection of productive early maturing cotton genotypes with improved fibre quality parameters. *Sci Horiz.* **27**(8):69-79.
- Malik W, Ashraf J, Iqbal MZ, *et al.* (2014) Molecular markers and cotton genetic improvement: current status and future prospects. *Sci World J.* **2014**:607091.
- Manan A, Zafar MM, Ren M, Khurshid M, Sahar A, Rehman A, *et al.* (2022) Genetic analysis of biochemical, fiber yield and quality traits of upland cotton under high-temperature. *Plant Prod Sci.* **25**(1):105-19.
- Manivannan A, Amal TC, Shanmugam A. (2025) Evaluation of genetic diversity of cotton genotypes using morphological and molecular markers and assessment of leaf hopper resistance. *South Afr J Bot.* **179**:206–215.
- Manonmani K, Mahalingam L, Malarvizhi D, Sritharan N, Premalatha N. (2019) Genetic variability, correlation and path analysis for seed cotton yield improvement in upland cotton (*Gossypium hirsutum* L.). *J Pharmacogn Phytochem.* **8**(4):1358-61.
- Mawblei C, Premalatha N, Rajeswari S, Manivannan A. (2022) Genetic variability, correlation and path analysis of upland cotton (*Gossypium hirsutum* L.) germplasm for seed cotton yield. *Electron J Plant Breed.* **13**(3):820-5.
- Meena RK, Parmar LD, Kishor R. (2022a) Genetic variability studies of yield and its component traits in upland cotton (*Gossypium hirsutum* L.). *Pharma Innov J.* **11**(5):380-4.
- Meena RK, Meena RK, Koli GK, Kumar D. (2022b) Insights of Genetic Divergence through Mahalanobis' D^2 Statistics among American Cotton (*Gossypium hirsutum* L.) Genotypes. *Biol Forum Int J.* **14**(2):790-5.
- Mei H, Zhu X, Zhang T. (2013) Favorable QTL alleles for yield and its components identified by association mapping in Chinese Upland cotton cultivars. *PLoS ONE.* **8**(12): e82193.

- Modi SS, Patel DH, Rajkumar BK, Parmar PR. (2020) Characterization of cotton germplasm through morphological characters and PCR based molecular markers. *J Pharmacogn Phytochem.* **9**(1):894-7.
- Monisha K, Premalatha N, Sakthivel N, Kumar M. (2018) Genetic variability and correlation studies in upland cotton (*Gossypium hirsutum* L.). *Electron J Plant Breed.* **9**(3):1053-9.
- Nadeem A, Shakeel A, Imtiaz M, *et al.* (2022) Genetic variability studies for yield and within boll yield components in cotton (*Gossypium hirsutum* L.). *Biol Clin Sci Res J.* **1**.
- Naik BM, Satish Y, Babu DR, *et al.* (2016) Genetic diversity analysis in American cotton (*Gossypium hirsutum* L.). *Electron J Plant Breed.* **7**(4):1002-6.
- Nawaz B, Sattar S, Malik TA. (2019) Genetic analysis of yield components and fiber quality parameters in upland cotton. *Int Multidiscip Res J.* **9**(9):13-9.
- Nazir A, Farooq J, Mahmood A, *et al.* (2013) Estimation of genetic diversity for CLCuV, earliness and fiber quality traits using various statistical procedures in different crosses of *Gossypium hirsutum* L. *Vestnik OrelGAU.* **43**(4):2-9.
- Nikhil PG, Nidagundi JM, Anusha HA. (2018) Correlation and path analysis studies of yield and fiber quality traits in cotton (*Gossypium hirsutum* L.). *J Pharmacogn Phytochem.* **7**(5):2596-9.
- Nisar S, Khan TM, Iqbal MA, Ullah R, Bhutta MA, Ahmad S, *et al.* (2022) Assessment of yield contributing quantitative traits in upland cotton (*Gossypium hirsutum*). *Sarhad J Agric.* **38**(1):353-5.
- Nivedha R, Rajeswari S, Premalatha N, *et al.* (2020) Inheritance study for lint colour in naturally brown coloured upland cotton (*Gossypium hirsutum* L.). *Electron J Plant Breed.* **11**(02):550-5.
- Pandiyan J, Ramalingam A, Pillai MA, Saravanan S. (2019) Genetic parameters of economic and fibre characters in upland cotton (*Gossypium hirsutum* L.). *Int J Curr Microbiol Appl Sci.* **8**(6):2084-90.
- Panse VG, Sukhatme PV. (1978) Statistical Methods for Agricultural Workers: Indian Council of Agricultural Research, New Delhi.
- Panse VG. (1957) Genetics of quantitative characters in relation to plant breeding. *Ind J Genet Plant Breed.* **7**(3):318-28.

- Patel HH, Patel KV, Suthar VL, Parmar RJ, Patel SK. (2023) Genetic variability exploration in upland cotton (*Gossypium hirsutum* L.) for yield and fiber traits. *J Pharmaceu Innov.* **12**(8):628-34.
- Patel RK, Desai ND, Shah HB. (2021) Relationship between boll traits and cotton yield under variable environments. *Cotton Science.* **33**(4):425–33.
- Patel VB, Shah NK. (2018) Role of Phenotypic Selection in Improving Economic Traits in Cotton. *Int J Plant Breed.* **12**(3):105-112.
- Patil SS, Deshmukh JD, Shinde MS. (2021) Genotypic variability and correlation analysis for fiber quality traits in upland cotton. *Int J Tropic Agric.* **39**(6):1787–94.
- Peakall R, Smouse PE. (2012) GenAlEx 6.5: Genetic analysis in Excel. Population genetic software for teaching and research-an update. *Bioinform.* **28**(19):2537–9.
- Pereira GS, Cazé ALR, Silva MG, Almeida VC, Magalhães FOC, Silva Filho JL, *et al.* (2015) Optimal use of SSR markers for varietal identification of upland cotton. *Pesquisa Agropecuária Brasileira.* **50**:571-81.
- Pettigrew WT. (2004) Physiological consequences of moisture deficit stress in cotton. *Crop Sci.* **44**(4):1265–1272.
- Pilgrim M, Willison S. (2009) Dive into Python 3. New York, NY, USA: A press.
- Praveen SKC, Raju S, Ebenezer BRR, Ajish M, Darling BS. (2019) Studies on genetic variability, heritability and genetic advance in cotton (*Gossypium hirsutum* L.). *Plant Archives.* **19**(1):934-7.
- Pritchard JK, Stephens M, Donnelly P. (2000) Inference of population structure using multilocus genotype data. *Genetics.* **155**(2):945–59.
- Pujer S, Siwach SS, Sangwan RS, *et al.* (2015) Genetic diversity in upland cotton (*Gossypium hirsutum* L.). *J Cotton Res Dev.* **29**(1):16-8.
- Pujer SK, Siwach SS, Deshmukh JD, Sangwan RS, Sangwan O. (2014) Genetic variability, correlation and path analysis in upland cotton (*Gossypium hirsutum* L.). *Electron J Plant Breed.* **5**(2):284-289.
- Python. (2023) Statistics - mathematical statistics functions. [cited 2023 Mar 5]. Available from: <https://docs.python.org/3/library/statistics.html>
- Qin H, Chen M, Yi X, Bie S, Zhang C, Zhang Y, *et al.* (2015) Identification of associated SSR markers for yield component and fiber quality traits based on frame map and upland cotton collections. *PloS one.* **10**(1): e0118073.

- R Core Team. (2025) R: A language and environment for statistical computing. Vienna, Austria: R Foundation for Statistical Computing.
- Rai P, Sangwan O. (2020) Correlation studies in elite lines of upland cotton (*Gossypium hirsutum* L.). *Electron J Plant Breed.* **11**(01):298-300.
- Raj DS, Patil RS. (2023) Genetic variability investigations on early maturity indicators and their inter-relationships in upland cotton (*Gossypium hirsutum* L.) cultivars. *Electron J Plant Breed.* **14**(4):1387-94.
- Raji AA. (2003) Assessment of genetic diversity and heterotic relationships in African improved and local cassava (*Manihot esculenta* Crantz) germplasm. Unpublished doctoral dissertation). University of Ibadan, Ibadan, Nigeria. 218p.
- Rakshit A, Rakshit S, Santhy V, Gotmare VP, Mohan P, Singh VV, *et al.* (2010) Evaluation of SSR markers for the assessment of genetic diversity and fingerprinting of *Gossypium hirsutum* accessions. *J Plant Biochem Biotechnol.* **19**(2):153-60.
- Rao PJM, Gopinath M. (2013) Association analysis of yield and fiber quality characters in upland cotton (*Gossypium hirsutum* L.). *Aust J Bas Appl Sci.* **7**(8):787-90.
- Rathinavel K. (2018) Principal component analysis with quantitative traits in extant cotton varieties (*Gossypium hirsutum* L.) and parental lines for diversity. *Curr Agril Res J.* **6**(1):54.
- Rathinavel K. (2019) Agro-morphological characterization and genetic diversity analysis of cotton germplasm (*Gossypium hirsutum* L.). *Int J Curr Microbiol Appl Sci.* **8**(2):2039-57.
- Rauf S, Shahzad K, Tehseen A, Khan TM. (2019) Genetic variability and correlation analysis of yield and fiber quality traits in upland cotton. *Int J Agric Biol.* **21**(3):559–66.
- Rauf SA, Khan TM, Sadaqat HA, Khan AI. (2004) Correlation and path coefficient analysis of yield components in cotton (*Gossypium hirsutum* L.). *Int J Agric Biol.* **6**(4):86-8.
- Raza H, Khan NU, Khan SA, Gul S, Latif A, Hussain I, *et al.* (2016) Genetic variability and correlation studies in F₄ populations of upland cotton. *JAPS: J Anim Plant Sci.* **26**(4):1048-55.
- Reddy BK, Reddy V. (2016) Genetic variability and heritability in upland cotton (*Gossypium hirsutum* L.). *Intl J Vet Sci Agric Res.* **4**(3):17-21.

- Reddy KB, Reddy VC, Ahamed ML, Naidu TCM, Srinivasarao V. (2015a) Correlation and path coefficient analysis in upland cotton (*Gossypium hirsutum* L.). *J Res ANGRAU*. **43**(1&2):25-35.
- Reddy KB, Reddy VC, Ahamed ML, Naidu TCM, Srinivasarao V. (2015b) Correlation and path coefficient analysis in upland cotton (*Gossypium hirsutum* L.). *Int J Pure Appl Biosci*. **3**(3):70-80.
- Reddy KR, Zhao D, Kakani VG. (2017). Effects of temperature and CO₂ on cotton growth, development and yield. *Agron J*. **109**(5):2082–2092.
- Reddy KVS, Balakrishna B, Reddy VC. (2019) Study of Cause and Effect Relationship among Yield, Yield Contributing and Quality Traits in Cotton (*Gossypium hirsutum* L.). *Intl J Vet Sci Agric Res*. **8**(6):1798-807.
- Rehman A, Farooq M. (2019). Morphology, Physiology and Ecology of Cotton. <https://doi.org/10.1002/9781119385523.ch2>
- Rehman A, Mustafa N, Du X, Azhar MT. (2020) Heritability and correlation analysis of morphological and yield traits in genetically modified cotton. *J Cotton Res*. **3**:1-9.
- Revanasiddayya, Nidagundi JM, Fakrudin B, Kuchanur P, Yogeesh LN, Hanchinal S, *et al.* (2024) Genetic diversity among coloured cotton genotypes in relation to their fibre colour and ploidy level based on SSR markers. *Czech J Genet Plant Breed*. **60**:12–24.
- Robinson HF, Comstock RE, Harvey PH. (1949) Estimate of heritability and the degree of dominance in corn. *Agron J*. **41**:353.
- Rudgers JA, Gardener MC. (2004) Extra floral nectar as a resource mediating multispecies interactions. *Ecology*. **85**(6):1495–502.
- Saeed F, Farooq J, Mahmood A, *et al.* (2014) Assessment of genetic diversity for Cotton leaf curl virus (CLCuD), fiber quality and some morphological traits using different statistical procedures in '*Gossypium hirsutum*' L. *Aust J Crop Sci*. **8**(3):442-7.
- Sahar A, Zafar MM, Razzaq A, Manan A, Haroon M, Sajid S, *et al.* (2021) Genetic variability for yield and fiber related traits in genetically modified cotton. *J Cotton Res*. **4**(1):1–10.
- Sainath, Patil BR, Patil RS, Nayak SN, Prasad MBP, Aishwarya B, *et al.* (2022) Correlation and path analysis studies for yield and yield attributes in recombinant

- inbred lines in cotton (*Gossypium hirsutum* L.). *Biol Forum - An Int J.* **14**(4):539-42.
- Salahuddin S, Abro S, Rehman A, Iqbal K. (2010) Correlation analysis of seed cotton yield with some quantitative traits in upland cotton (*Gossypium hirsutum* L.). *Pak J Bot.* **42**(6):3799-805.
- Salama EA, Farid MA, El-Mahalawy YA, El-Akheder AAA, Aboshosha AA, Fayed AM, *et al.* (2024) Exploring agro-morphological and fiber traits diversity in cotton (*G. barbadense* L.). *BMC Plant Biol.* **24**(1):403.
- Salgotra RK, Chauhan BS. (2023) Genetic Diversity, Conservation, and Utilization of Plant Genetic Resources. *Genes (Basel).* **14**(1):174.
- Santhy V, Meshram M, Santosh HB, Kranthi KR. (2019) Molecular diversity analysis and DNA fingerprinting of cotton varieties of India. *Ind J Genet Plant Breed.* **79**(04):719-25.
- Saraswat H, Sonika, Sanwan O, *et al.* (2022) Characterization of upland Cotton (*Gossypium*) Genotypes as per PPV&FR Guidelines via Important Morphological Traits. *Biological Forum- An Int J.* **14**(3):1461-8.
- Sarode D, Sharma K, Shinde N, Gaikwad A, Chavhan R, Pimpale P. (2021) Molecular characterization of cotton (*Gossypium hirsutum* L.) genotypes using SSR markers for fiber quality traits. *Int J Curr Microbiol App Sci.* **10**(03):1748-59.
- Sarwar G, Nazir A, Rizwan M, *et al.* (2021a) Assessment of genetic diversity of cotton cultivars by using correlation and principal component analysis for CLCuD tolerance, yield and quality traits. *J Agric Res.* **59**(1):1-5.
- Sarwar G, Nazir A, Rizwan M, *et al.* (2021b) Genetic diversity among cotton genotypes for earliness, yield and fiber quality traits using correlation, principal component and cluster analyses. *Sarhad J Agril.* **37**(1):307-14.
- Sarwar G, Nazir A, Shahzadi E, Mahmood A. (2020) Assessment of genetic diversity in cotton genotypes for earliness, clcu disease tolerance, fiber quality and yield related traits. *J Agril Res.* **58**(3):165-70.
- Selvaraj S, Rajeswari S, Premalatha N, Kalaimagal T, Muthuswami M, Jeyakumar P, *et al.* (2023) Association and path analysis studies for yield contributing and fibre quality traits in the F₁ population of *Gossypium hirsutum* L. *Environ Conserv J.* **24**(2):70-6.

- Seyed MSY, Sofalian O, Asghari A, Sedghi M, Zangi M. (2022a) Selection and introduction of high yield and early cotton cultivars from advanced cultivars in Ardabil province. *Iran J Cotton Res.* **9**(2):165-77.
- Seyyed MSY, Sofalian O, Asghari A, Sedghi M, Zanghi MR. (2022b) Evaluation of earliness and yield-related traits in advanced cotton cultivars (*Gossypium hirsutum* L.). *Prog Chem Biochem Res.* **5**(3):301-16.
- Shabbir RH, Bashir QA, Shakeel A, Khan MM, Farooq J, Fiaz S, *et al.* (2016) Genetic divergence assessment in upland cotton (*Gossypium hirsutum* L.) using various statistical tools. *J Glob Innov Agril Social Sci.* **4**(2):62-9.
- Shah SA, Khan SJ, Ullahand K, *et al.* (2018) Genetic diversity in cotton germplasm using multivariate analysis. *Sarhad J Agril.* **34**(1):130-5.
- Shaheen M, Abdul-Rauf H, Taj MA, *et al.* (2021) Path analysis based on genetic association of yield components and insect's pest in upland cotton varieties. *PloS One.* **16**(12): e0260971.
- Shahzad K, Li X, Qi T, Guo L, Tang H, Zhang X, *et al.* (2019) Genetic analysis of yield and fiber quality traits in upland cotton (*Gossypium hirsutum* L.) cultivated in different ecological regions of China. *J Cotton Res.* **2**(1):14.
- Shakeel A, Azhar MT, Ali I, *et al.* (2018) Genetic diversity for seed cotton yield parameters, protein and oil contents among various Bt. cotton cultivars. *Int J Biosci.* **12**(1):242-51.
- Shakeel A, Talib I, Rashid M, *et al.* (2015) Genetic diversity among upland cotton genotypes for quality and yield related traits. *Pak J Agric Sci.* **52**(1):73-7.
- Shang L, Liang Q, Wang Y, Wang X, Wang K, Abdulkarimov A, *et al.* (2016) Identification of QTLs for fiber quality and yield traits using a high-density genetic map in upland cotton (*Gossypium hirsutum* L.). *Theo Appl Genet.* **129**(4):755–68.
- Sharma D, Kumari M, Sandeep S, Thakur N. (2023) Correlation and Path Coefficient Analysis for Yield and Component Traits in Pea (*Pisum sativum* L.). *Bd J Bot.* **52**(4):907-14.
- Sharma JR. (2006) Statistical and biometrical techniques in plant breeding. New Age International. 432.
- Shi J, Zhang Y, Wang N, Xu Q, Huo F, Liu X, *et al.* (2023) Genetic diversity and population structure analysis of upland cotton (*Gossypium hirsutum* L.)

- germplasm in China based on SSR markers. *Genetic Resour Crop Evol.* **70**(8):2671–82.
- Shinde CS, Shitole LS, Bhor TJ, Jadhav DP, Nimbalkar RD, Wagh RS. (2025) Molecular characterization of cotton genotypes using PCR based molecular markers. *Vegetos.* **38**(1):1-11.
- Shoukat S, Anwar M, Ali S, Begum S, Aqeel M, Zia MA, *et al.* (2020) Estimation of morphological and molecular diversity of seventy-two advanced Pakistani cotton genotypes using simple sequence repeats. *Appl Ecol Environ Res.* **18**(2):2043-56.
- Shruti, Sowmya HC, Nidagundi JM, Lokesh R, Arunkumar B, Murthy SM. (2020) Correlation and Path Coefficient Analysis for Seed Cotton Yield, Yield Attributing and Fiber Quality Traits in Cotton (*Gossypium hirsutum* L.). *Int J Curr Microbiol Appl Sci.* **9**(02):200-7.
- Shruti, Sowmya HC, Nidagundi JM, Lokesh R, Arunkumar B, Murthy SM. (2019) Genetic variability studies for yield, yield attributing and fiber quality traits in cotton (*Gossypium hirsutum* L.). *Int J Curr Microbiol Appl Scie.* **8**(10):2677-87.
- Singh A, Kumar V. (2020) Impact of maturity duration on cotton yield and quality traits: A review. *J Plant Breed Crop Sci.* **12**(3):47–56.
- Singh P, Narayanan SS. (1993) Biometrical techniques in plant breeding. Kalyani Publishers.
- Singh R, Sharma P, Singh J. (2020) Plant height variability and its effect on yield stability in upland cotton. *Agril Sci J.* **15**(2):110–8.
- Singh RK, Chaudhary BD. (1985) Biometrical methods in Quantitative Genetic Analysis. New Delhi-Ludhiana, India: Kalyani Publishers. 56.
- Sirisha ABM, Ahamed ML, Kumar PR, Kumari SR, Rao VS. (2019) Heterosis studies for fibre quality traits in upland cotton (*Gossypium hirsutum* L.). *The Andhra Agril J.* **66**(2):293-5.
- Smith CW, Stephens JG. (2011) Cotton: Origin, History, Technology, and Production. John Wiley & Sons.
- Smith OB, Jones AC. (2019) Quantitative Genetics of Fiber Traits in Upland Cotton. *J Crop Improv.* **33**(2):200-215.
- Song MZ, Fan SL, Yuan RH, Pang CY, Yu SX. (2012) Genetic analysis of earliness traits in short season cotton (*Gossypium hirsutum* L.). *J Integr Agric.* **11**(12):1968-75.

- Soomro AW, Channa AR, Ahsan MZ, Majidano MS, Panhwar FH. (2015) Assessing of Plant trait for predicting earliness in upland cotton (*Gossypium hirsutum*, L.). *American-Eurasian J Agric & Environ Sci.* **15**(1):48-52.
- Srinivas B, Bhadru D, Rao MV. (2015) Correlation and path coefficient analysis for seed cotton yield and its components in American cotton (*Gossypium hirsutum* L.). *Agril Sci Digest-A Res J.* **35**(1):13-8.
- Statistix 10. (2025) Data analysis software for researchers.
- Subalakhshmi VKI, Rajeswari S, Premalatha N, Thirukumaran K, Boopathi NM. (2023) Association analysis for economic traits in upland cotton (*Gossypium hirsutum* L.). *Ind J Agril Res.* **57**(2):143-9.
- Subhashini S, Rajeswari S, Premalatha N, Kalaimagal T, Muthuswami M, Jeyakumar P, *et al.* (2023) Association and path analysis studies for yield contributing and fibre quality traits in the F₁ population of *Gossypium hirsutum* L. *Environ Conserv J.* **24**(2):70-76
- Sudeepthi K, Jyothula DP, Suneetha Y. (2018) Heterosis and combining ability studies for yield and yield component traits in rice (*Oryza sativa* L.). *Int J Curr Microbiol App Sci.* **7**(10):1205-11.
- Sun Z, Wang X, Liu Z, Gu Q, Zhang Y, Li Z, *et al.* (2017) Genome-wide association study discovered genetic variation and candidate genes of fibre quality traits in (*Gossypium hirsutum* L.). *Plant Biotechnol J.* **15**(8):982-96.
- Sun Z, Wang X, Liu Z, Gu Q, Zhang Y, Li Z, *et al.* (2019) Evaluation of the genetic diversity of fibre quality traits in upland cotton (*Gossypium hirsutum* L.) inferred from phenotypic variations. *J Cotton Res.* **2**(1):22.
- Sundar R, Patil SS, Ranganath HM, Srivalli P, Manjula SM, Pranesh KJ. (2014) Genetic diversity analysis in cotton (*Gossypium hirsutum* L.) based on morphological traits and microsatellite markers. *Int J Bas Appl Biol.* **1**(1):2349-5820.
- Sunilkumar V, Campbell LM, Puckhaber L, Stipanovic RD, Rathore KS. (2017) Engineering cottonseed for use in human nutrition by tissue-specific reduction of toxic gossypol. *Proceedings of the Nation Academy Sciences.* **103**(48):18054–9.
- Swarup S, Cargill EJ, Crosby K, Flagel L, Kniskern J, Glenn KC. (2021) Genetic diversity is indispensable for plant breeding to improve crops. *Crop Science.* **61**(2):839-52.

- Swarup V, Chaugle BS. (1962) Studies on genetic variability in sorghum. Phenotypic variation and heritable component in some quantitative characters contributing towards yield. *Ind J Genet Plant Breed.* **22**(1):31-6.
- Tabib MFAI. (2023) Ecology and economics of cotton in Bangladesh. *Bd Textile Today.*
- Teklewold A, Jayaramaiah H, Jagadeesh BN. (2000) Correlations and path analysis of physio-morphological characters of sunflower (*Helianthus annuus* L.) as related to breeding method. *Helia.* **23**(32):105-14.
- Tu JL, Zhang MJ, Wang XQ, Zhang X, Lin Z. (2014) Genetic dissection of upland cotton (*Gossypium hirsutum*) cultivars developed in Hubei Province by mapped SSRs. *Genet Mol Res.* **13**(782):90.
- Tyagi P, Gore MA, Bowman DT, Campbell BT, Udall JA, Kuraparthi V. (2014) Genetic diversity and population structure in the US upland cotton (*Gossypium hirsutum* L.). *Theor Appl Genet.* **127**(2):283–95.
- Ullah I, Iram A, Iqbal M, Nawaz M, Hasni S, Jamil S. (2012) Genetic diversity analysis of Bt cotton genotypes in Pakistan using simple sequence repeat markers. *Genet Mol Res.* **11**(1):597-605.
- Ullah Q, Ahmad MZ, Ullah K, *et al.* (2020) Investigation of cotton germplasm for genetic divergence regarding yield related trait using principal component analysis. *Res Square.* 1-12.
- Ulloa M. (2006) Heritability and correlations of agronomic and fiber traits in an okra-leaf upland cotton population. *Crop Sci.* **46**(4):1508-14.
- USDA Foreign Agricultural Service (FAS). (2024) Bangladesh: Cotton and Products Update (GAIN Report BG2024-0012). Washington, DC: USDA.
- USDA Foreign Agricultural Service (FAS). (2025) Global Agricultural Information Network (GAIN)- Cotton and Products Outlook MY 2025/26. Washington, DC: USDA.
- U.S. Cotton Fiber Chart. (2023/2024) Ratings of Fiber Properties and Grades of U.S. Cotton. Cotton Incorporated.
- Vadodaria H, Patidar D, Kumar S, Patel DA, Patel H. (2024) Characterization of desi cotton (*Gossypium arboreum*) germplasm through morpho-biochemical and fiber traits and exploring molecular marker based diversity along with allied (un) cultivated *Gossypium* species. *CABI Agric Biosci.* **5**(1):111.

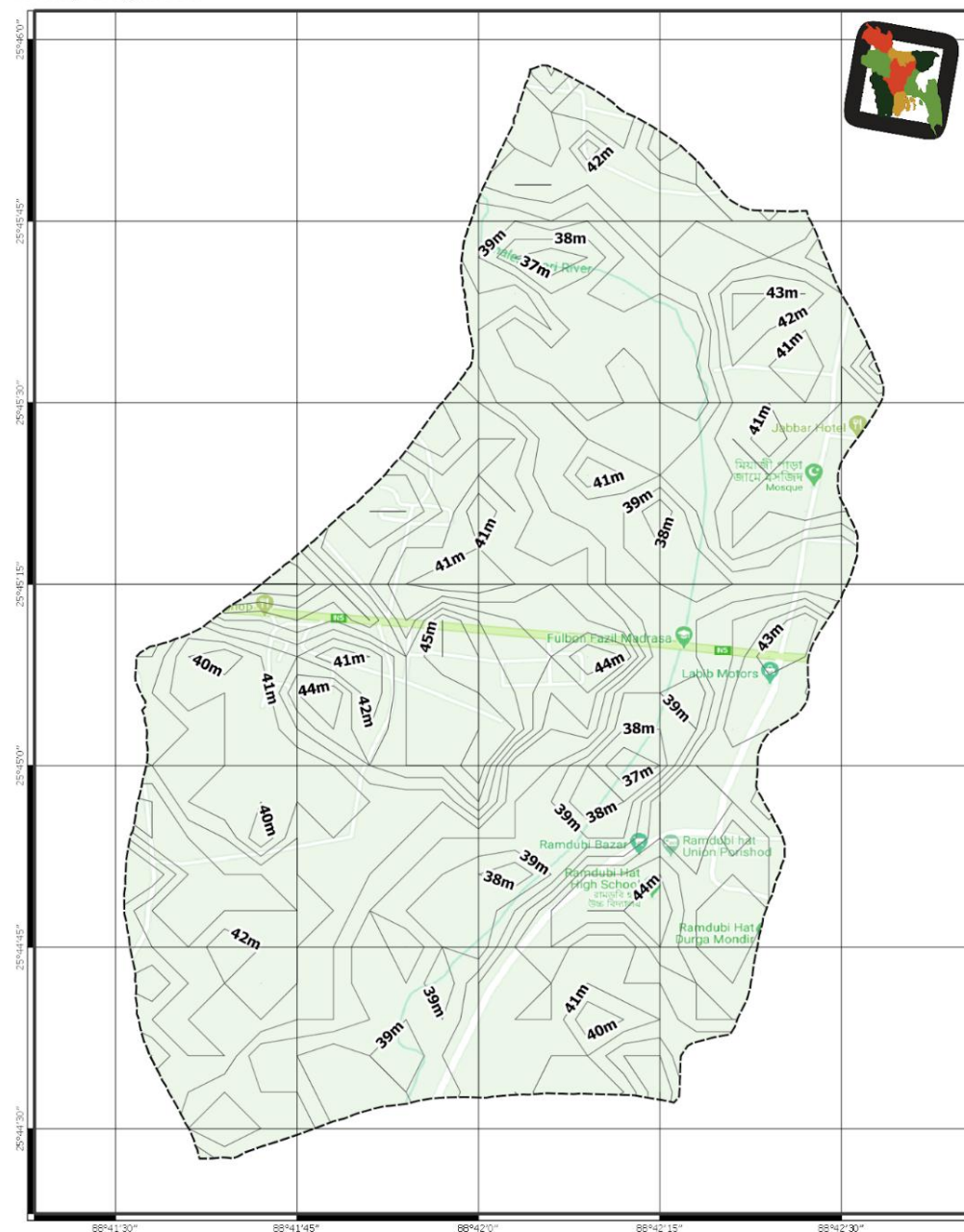
- Vishnuvardhan KM, Reddy BV, Lakshmikalyani D, *et al.* (2023) Multivariate Analysis in Upland Cotton (*Gossypium hirsutum* L.) using Principal Component and Cluster Analysis. *Eco Env & Cons.* **29**:224-30.
- Wang Y, Yang C, Liu G, Zhang G, Ban Q. (2007) Microarray and suppression subtractive hybridization analyses of gene expression in *Puccinellia tenuiflora* after exposure to NaHCO₃. *Plant Sci.* **173**(3):309-20.
- Ward Jr JH. (1963) Hierarchical grouping to optimize an objective function. *J Am Stat Assoc.* **58**(301):236-44.
- Wegier A, Piñeyro-nelson A, Alarcón J, Gálvez-mariscal A, Álvarez-Buylla ER, Piñero D. (2011) Recent long-distance transgene flow into wild populations conforms to historical patterns of gene flow in cotton (*Gossypium hirsutum*) at its centre of origin. *Mol Ecol.* **20**(19):4182-94.
- Wells R, Meredith WR. (1984) Comparative growth of obsolete and modern cotton cultivars: I. Vegetative and reproductive dry matter partitioning. *Crop Sci.* **24**(5), 858–862.
- Wendel JF, Brubaker CL, Percival AE. (1992) Genetic Diversity in *Gossypium hirsutum* and the Origin of Upland Cotton. *Ame J Bot.* **79**:1291-310.
- Wendel JF, Cronn RC. (2003) Polyploidy and the evolutionary history of cotton. *Adv Agron.* **87**:139-86.
- Wendel JF, Grover CE. (2015) Taxonomy and evolution of the cotton genus, *Gossypium*. Cotton, Agronomy Monograph **57**. ASA, CSSA, and SSSA.
- Wendel JF. (1989) "New World Tetraploid Cottons Contain Old World Cytoplasm," *Proceedings of the National Academy of Science (U.S.A.)*. **86**:4132-6.
- Wiley Online Library. (2009) Biodegradable polymers in the circular plastics economy.
- Wright S. (1921) Correlation and causation. *J Agril Res.* **20**(7):557.
- Wu Y, Huang L, Zhou D, Fu X, Li C, Wei S, *et al.* (2020a) Development and application of perfect SSR markers in cotton. *J Cotton Res.* **3**(1):21.
- Xu CC, Zhang QR, Hu L, Wang RM, Shi Q, Chen J, *et al.* (2020) Seed soaking with sodium silicate primes salt tolerance in rice (*Oryza sativa* L.) seedlings without any negative effect on growth. *Appl Ecol Environ Res.* **18**(4):5425–36.
- Yaqoob M, Fiaz S, Ijaz B. (2016) Correlation analysis for yield and fiber quality traits in upland cotton. *Commun Plant Sci.* **6**(3-4):55-60.
- Yaqoob M, Fiaz S, Ijaz B. (2020) Heritability and correlation analysis of morphological and yield traits in genetically modified cotton. *J Cotton Res.* **3**(1):1–8.

- Yasar M. (2023) Yield and fiber quality traits of cotton (*Gossypium hirsutum* L.) cultivars analyzed by biplot method. *Journal of King Saud University - Science*. **35**:102632.
- Yuzhen WU, Longyu H, Dayun Z, Xiaoqiong Z, Chao L, Shoujun W, PENG Jun P and Meng K. (2020) Development and application of perfect SSR markers in cotton. *J Cotton Res*. 3:21.
- Zafar MM, Zhang Y, Farooq MA, *et al.* (2022) Biochemical and associated agronomic traits in *Gossypium hirsutum* L. under high temperature stress. *Agron*. **12**(6):1310.
- Zhang J, Guo W, Zhang T. (2002) Molecular linkage map of allotetraploid cotton (*Gossypium hirsutum* L. x *Gossypium barbadense* L.) with a haploid population. *TAG- Theo Appl Genet*. **105**(8):1166–74.
- Zhang YC, Kuang M, Yang WH, Xu HX, Zhou DY, Wang YQ, *et al.* (2013) Construction of a primary DNA fingerprint database for cotton cultivars. *Genet Mol Res*. **12**(2):1897–906.
- Zhang Z, Li X, Yu R, Han Y, Zhang G. (2019) Correlation and path coefficient analysis of yield and fiber quality traits in upland cotton (*Gossypium hirsutum* L.). *J Cotton Res*. **2**(1):1–8.
- Zhao L, Wang Y, Li X. (2021) Early flowering in cotton: Implications for yield and fiber quality. *Field Crops Res*. **265**:10807.

APPENDICES

Appendix I: Map of the experimental area

Mouza: Sadarpur
Union: Sundarban
Upazila: Dinajpur Sadar
District: DINAJPUR
Country: Bangladesh



Data Source: The Bangladesh Network
URL: <https://www.thebangladesh.net>

Created on: April, 2020.
Vector Source: The Bangladesh Network ©.

Appendix II: Weather data of the experimental site during the period from July-December, 2021

Month	Relative Humidity (%)	Temperature		Total Rainfall (mm)
		Minimum (°C)	Maximum (°C)	
July-2021	79	27	32	301.61
August-2021	81	26	31	292.73
September-2021	79	26	32	204.56
October-2021	72	25	32	185.48
November-2021	56	19	30	6.0
December-2021	55	16	26	134.75

Source: www.worldweatheronline.com

Appendix III: Weather data of the experimental site during the period from July-December, 2022

Month	Relative Humidity (%)	Temperature		Total Rainfall (mm)
		Minimum (°C)	Maximum (°C)	
July-2022	73	27	33	141.48
August-2022	74	27	33	149.52
September-2022	79	26	32	397.0
October-2022	75	23	31	184.57
November-2022	59	19	30	0.50
December-2022	48	16	28	1.40

Source: www.worldweatheronline.com

Appendix IV: Weather data of the experimental site during the period from July-December, 2023

Month	Relative Humidity (%)	Temperature		Total Rainfall (mm)
		Minimum (°C)	Maximum (°C)	
July-2023	77	27	33	225.70
August-2023	80	26	32	448.05
September-2023	78	26	32	265.50
October-2023	70	24	31	140.09
November-2023	59	21	30	15.58
December-2023	57	18	27	6.73

Source: www.worldweatheronline.com

Appendix V: The soil nutrient composition of the experimental site

Physical and chemical properties	Value
Sand (%)	65
Silt (%)	30
Clay (%)	5
Textural Class	Sandy loam
pH	5.86
AEZ	03
Latitude	25° 45' 19" N
Longitude	88° 41' 32" E
Altitude	34.5 m
Organic Carbon (%)	0.84
Organic Matter (%)	1.44
Total Nitrogen (%)	0.07
Phosphorus (µg/g)	70.46
Potassium (meq/100g)	0.43
Calcium (meq/100g)	4.19
Magnesium (meq/100)	0.67
Sulphur (µg/g)	8.33
Zinc (µg/g)	14.37
Coper (µg/g)	0.85
Iron (µg/g)	48.31
Manganese (µg/g)	6.01
Boron (µg/g)	0.14

Here, N= North, E= East, m= meter, µg= micro gram, g= gram, meq= mili equivalent

Source: Soil Resources Development Institute (SRDI), Nashipur, Dinajpur-5200.