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# Assessing genetic variation in *Gossypium barbadense* L. germplasm based on fibre characters

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## Abstract

**Background** *Gossypium barbadense* L. has specific fibre in terms of its length, strength, and fineness, and known as extra-long staple (ELS) cotton, Sea-Island cotton, or Egyptian cotton. Narrow genetic base with less genetic variability is observed in *G. barbadense* germplasm. Hence, this study was aimed to evaluate the genetic variability present in 108 germplasm accessions of *G. barbadense* and to identify the superior genotypes based on the fibre traits.

**Results** We evaluated 108 accessions for five fibre quality traits along with three checks in augmented block design. All fibre traits showed significant differences among genotypes, indicating that there is genetic potential for improvement. Fibre strength and micronaire (MIC) showed high phenotypic and genotypic coefficients of variation. High heritability combined with high genetic advance as percentage of mean (GAM) was recorded for fibre length, strength, and micronaire. Fibre strength and fibre length were significantly correlated with each other, while both showed negative correlation with micronaire. Principal component analysis and Biplot analysis showed that uniformity index discriminated all the genotypes in higher level, while fibre length and strength were medium in discrimination power. Biplot revealed genotypes DB 16, EC959191, GSB 39, ARBB 20, 5746U, EA 203, and EA 201 were genetically diverse. Hierarchical cluster analysis based on unweighted paired group method using arithmetic average (UPGMA) grouped the genotypes into four clusters, with each cluster consisting of 4, 18, 48, and 38 genotypes, respectively.

**Conclusion** Among the genotypes, 34 for fibre length ( $> 35$  mm), 18 for fibre strength ( $> 40.4$  g·tex<sup>-1</sup>) and 66 for micronaire (3.7–4.2, A grade) were identified as potential accessions based on their superiority. The superior fibre genotypes identified in this study are potential lines for the ELS cotton breeding program.

**Keywords** Characterization, Diversity, Extra-long staple cotton, Fibre quality traits, Germplasm, *Gossypium barbadense*

## Background

Cotton is the major source of raw material in terms of natural fibre used in textile industries. Cotton fibre is formed from an epidermal cell growth as an extension of single cell from the seed surface; hence, we call it as

surface fibre or seed fibre. Cotton fibre is the purest cellulose form and the most abundant polymer among natural fibres, which accounts for 90% of cellulose. The cellulose found in cotton fibre is considered as having the highest molecular weight and structural order of any plant fibres (Hsieh 2007). Cotton belongs to the genera *Gossypium* of Malvaceae family. The genus *Gossypium* has seven tetraploid species and around 45 diploid species in different ecological niches from tropical to semitropical to arid regions (Wang et al. 2022). A, B, C, D, E, F, G, and K genomes are found among them (Wendel et al. 2012). Among tetraploid species of *Gossypium*, two of them are

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cultivated species, upland cotton or American cotton (*G. hirsutum* L.,  $2n=4x=52$ ,  $A_1A_1D_1D_1$  genome), which has been widely cultivated to the extent of 90% cotton planting area. Egyptian cotton or Sea-Island cotton (*G. barbadense* L.,  $2n=4x=52$ ,  $A_2A_2D_2D_2$  genome) occupies 5%-8% planting area. While in diploid species, two are cultivated, Asiatic cotton or *desi* cotton (*G. arboreum* L.,  $2n=26$ ,  $A_2A_2$  genome) has the area of 2%-5%; and Levant cotton (*G. herbaceum* L.,  $2n=26$ ,  $A_1A_1$  genome) which mainly grows in marginal and insignificant areas in Indian subcontinent. Out of the four cultivated species, *G. barbadense*, often known as extra-long staple (ELS) cotton (Egyptian cotton, or Pima cotton, or Sea-Island cotton) is renowned for its outstanding fibre characteristics. It has been cultivated in India, China, Egypt, USA, Peru, Israel, Tajikistan, Turkmenistan, and Uzbekistan, etc. Compared with *G. hirsutum*, *G. barbadense* is low in yield, and poor in adaptable, susceptible for sucking pests, and difficult to pick manually as it has prominent pointed edges on the boll. However, its fibre is soft, very long, lustrous, and strong, which accounts for greater number of counts while spinning, and it is being used for producing premium garments and textile materials. ELS fibre is favored for spinning the stronger and more silky yarns that can be made into upscale cotton garments (Avci et al. 2013). Hence, it offers potential for development of the superior fibre and premium textile produces. Therefore, studies on fibre quality characters of *G. barbadense* is gaining momentum at present. An ideal fibre is considered as white as frozen vapor, sturdy like iron, soft like silk, and stretches like wool; in total cotton fibre is felt like woven wind (Bradov and Davidonis 2000). In real sense, obtaining such ideal fibre seems not feasible. However, considering all other *Gossypium* species, *G. barbadense* possesses near to such an ideal cotton.

Quality improvement is the prime focus for cotton breeders since fibre quality have a direct effect on yield and ever since enhancements in spinning technology have redoubled the demand for top-grade fibre (Wendel and Cronn 2003). Fibre quality is decided by the mixture of attributes including length, strength, fineness,

elasticity, uniformity index (UI), short fibre content, color, reflectance, naps, and spinning consistency, etc. (Hake et al. 1996). In the manufacture of textiles, these properties are important for spinning, weaving, and dyeing (Asif et al. 2008). Length of fibre has a direct relationship with yarn quality, strength, and spinning efficiency. Strength of fibre is considered as a crucial factor in determining the spinning. The fibre processing and dyeing uniformity are mainly influenced by micronaire (MIC), a gauge of fibre fineness and maturity. In addition, in the textile sector, fibre uniformity and elongation are of great importance, too. They are closely linked to the advanced processes of spinning and weaving which convert fibre into textiles (McCarty et al. 2006).

In terms of fibre characteristics, existing commercial cotton varieties are characterized by limited variability. Furthermore, the environment has a very strong influence on fibre quality parameters due to its heterogeneous nature of inheritance, which makes it difficult to breed improved fibre (Percy et al. 2006). The presence of genetic variation and its correlation with traits are particularly important to breeding programmes. In order to introduce a successful breeding programme for the improvement of genotypes with better fibre quality, it is crucial that genetic variability and correlation among fibre properties of cotton are studied and the existing germplasm pools are screened for further varietal improvements (Ulloa 2006). Genetic improvement for enhancing fibre quality traits is the prime objective of many cotton breeding programmes. The aim of our study is to evaluate the genetic variation among the 108 *G. barbadense* accessions for fibre quality traits, to analyse the association among fibre traits, and to identify the superior accessions for future breeding programmes.

## Results

### Analysis of components of variance

The current study showed a wide variation for the fibre traits analysed (Table 1). UHML (Upper half mean length) ranged from 24.3 to 40.3 mm with a mean of 33.94 mm. The trait tenacity or tensile strength or fibre

**Table 1** Mean, range and variance components of *G. barbadense* germplasm for fibre traits

Characters	Mean	Range	PV	GV	PCV/%	GCV/%	$H^2$ /%	GA	GAM/%
UHML	33.94 mm	24.3-40.3 mm	8.80	5.30	8.74	6.78	60.22	3.68	10.84
UI	85.84%	81%-89%	2.15	1.70	1.71	1.52	79.10	2.39	2.78
MIC	4.11	3.2-5.9	0.26	0.20	12.34	10.80	76.62	0.80	19.47
STR	36.49 g·tex <sup>-1</sup>	23.2-45.6 g·tex <sup>-1</sup>	16.44	11.87	11.11	9.44	72.20	6.03	16.53
EL	6.75%	6.1%-7.0%	0.03	0.02	2.63	2.17	68.25	0.25	3.70

UHML: Upper half mean length, UI: Uniformity index, MIC: Micronaire, STR: Strength, EL: Elongation percentage, PV: Phenotypic variance, GV: Genotypic variance, PCV: Phenotypic coefficient of variance, GCV: Genotypic coefficient of variance,  $H^2$ : Heritability, GA: Genetic advance, GAM: Genetic advance as percentage of mean

strength varied from 23.2 g-tex<sup>-1</sup> to 45.6 g-tex<sup>-1</sup> with a mean strength of 36.49 g-tex<sup>-1</sup>. In case of micronaire, the range was 3.2 to 5.9 with a mean value of 4.11. However, UI showed a narrow range from 81% to 89% with a mean value of 85.84%, and fibre elongation percentage showed the range of 6.1%–7.0% with a mean value of 6.75%.

Variance caused by phenotype and genotype indicates the heritable variation based on phenotypic expression of particular traits. In the current study, it was found that PV was higher than GV for the five fibre traits. Elongation percentage showed a low PV (0.03), but the PV of fibre strength was high (16.44). The same trend was observed for GV, as low in elongation percentage (0.02) but high in strength (11.87). High PCV and GCV were observed for strength and micronaire. While, other traits were observed medium to lower values. UI showed low PCV (1.17%), but PCV of micronaire was high (12.34%). The same trend was observed for GCV as low in UI (1.52%) but high in micronaire (10.80%). It was observed all the five fibre traits exhibited a higher PCV value than GCV, respectively (Table 1).

#### Estimates of heritability in broad sense ( $H^2$ ) and genetic advance (GA)

High heritability was observed in all fibre traits (60%–80%). The process of selection could be useful for characters with high heritability value. In case of GA, elongation percentage was found to be low (0.25), but fibre strength was high (6.03). As for GAM, micronaire was found to be high (19.47%), but UI was low (2.78%) (Table 1).

#### Principal component analysis (PCA)

Among the principal components (PCs), three out of five PCs were found to have eigen values more than 1.00. These PCs contributed for more than 90% variability of the traits (Table 2). PC1 was mainly explained by the fibre strength (0.810), fibre length (0.568), and UI (0.133). PC2 was mainly related with UI (0.956). PC3 was mainly associated with fibre length (0.773) and UI (0.249). PC4 was mainly related with micronaire (0.993), while PC5

was mainly correlated with fibre elongation percentage (0.998) (Table 3). As per PCA, the greatest variation was observed in fibre strength (48.8%), moderate variation in fibre length (31.8%) and UI (14.8%). However, smaller variation was found in micronaire (4.0%) and minimal variation in elongation percentage (0.6%) (Table 4).

#### Biplot analysis

PCA is the multivariate linear regression model (Pearson 1901) used in this analysis. PCA evaluates the results derived from the correlation analysis. Here, measured values are used to build a multidimensional data set, which is projected on vector plane called biplot. Biplot is a scatter plot that portrays the relationship among observed measures and dependent variables in terms of PCs (ter Braak 1983). In a PCA-based biplot, spots seen on the plane are the projected observations and vectors are the projected variables. Biplot was drawn using first two principal components namely PC1 and PC2, genotypes are scattered on the plane. Variables used for discerning the genotypes are traits of the germplasm, each trait been depicted as vectors. Vector length is proportional to the ability to discriminate genotypes at the biplot level. The vector with longer length has the greater discriminating power among the genotypes, and *vice versa*. UI has a longer vector as it discriminates the genotypes in higher level, while fibre length and strength were medium in discrimination power and almost in the

**Table 3** Eigen vector for fibre traits of *G. barbadense* accessions

Characters	PC1	PC2	PC3	PC4	PC5
UHML	0.568	-0.281	0.773	0.006	-0.026
UI	0.133	0.956	0.249	-0.071	-0.020
MIC	-0.060	0.066	0.059	0.993	-0.053
STR	0.810	0.044	-0.579	0.081	0.014
EL	0.003	0.015	0.037	0.051	0.998

UHML: Upper half mean length, UI: Uniformity index, MIC: Micronaire, STR: Strength, EL: Elongation percentage

**Table 4** Variability by different fiber traits of *G. barbadense* accessions

Characters	Variability contribution/%
UHML	31.8
UI	14.8
MIC	4.0
STR	48.8
EL	0.6

UHML: Upper half mean length, UI: Uniformity index, MIC: Micronaire, STR: Strength, EL: Elongation percentage

**Table 2** Principal component analysis (PCA) of fiber traits of *G. barbadense* accessions

PC	Eigen value	Proportion of variation/%
1	24.14	67.92
2	1.77	16.46
3	1.33	14.86
4	0.18	0.65
5	0.03	0.10

In the past several years, cotton production and consumption in India have been changing due to increased exports that outweigh domestic consumption. Competition has intensified and cotton production objectives have been altered because of shifting towards foreign markets, in parallel with switching to ring spinning over rotor spinning. Textile industry requires superior cotton that can endure all spinning processes to produce finer fabrics. In terms of fibre quality, *G. barbadense* is the best one as it is known for its ELS fibre and high strength, luster and fineness. The intrinsic negative association between fibre quality and yield in cotton drives the development of premium fibre. The majority of cotton germplasm has been derived from a limited genetic variability with narrow base due to limited cross-pollination or seed mixing. A single population underwent natural selection,



**Table 5** Inter correlation among fiber traits of *G. barbadense* accessions

Characters	UHML	UI	MIC	STR	EL
UHML	1.000				
UI	0.373 <sup>a</sup>	1.000			
MIC	-0.521 <sup>a</sup>	-0.094	1.000		
STR	0.879 <sup>a</sup>	0.423 <sup>a</sup>	-0.562 <sup>a</sup>	1.000	
EL	0.122	0.174	0.860 <sup>a</sup>	0.034	1.000

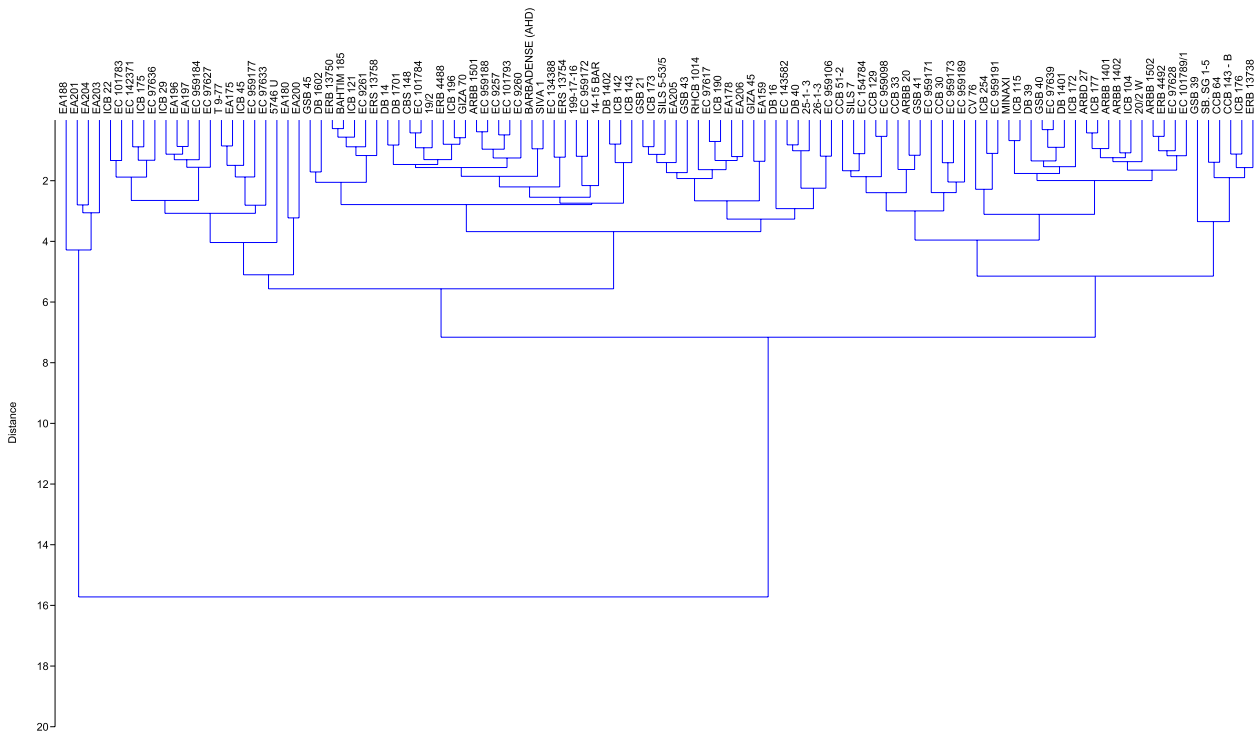
<sup>a</sup> Significant difference at 5%; UHML: Upper half mean length, UI: Uniformity index, MIC: Micronaire, STR: Strength, EL: Elongation percentage

which led to developing of modern cultivars with a limited genetic base. The screening of germplasm is necessary to identify such kinds of genotypes for further breeding programmes (McCouch et al. 2013). Many studies have been conducted on the genetics of fibre quality traits and fibre improvement in *G. hirsutum*, however, fewer studies have been done in *G. barbadense* (Wang et al. 2023). By considering all the facts, this study was envisaged to characterize *G. barbadense* germplasm with respect to fibre quality traits. Genotypes showed wide variation for all fibre traits in this study.

Sivasubramaniam and Madhav Menon (1973) classified PCV and GCV values into three classes: low (< 10%), medium (10%-20%), and high (> 20%). In this study,

except fibre strength and micronaire, all other traits (fibre length, UI, elongation percentage) fell in the class of low PCV and GCV, which implied that these characters are more influenced by the environment. Higher values of PCV compared with GCV showed that environmental effect is greater in the manifestation of these traits (Gadissa et al. 2019). Singh (2001) grouped heritability into three groups, low of less than 30%, medium in the range of 30%-70%, and high of more than 70%. Moderately-high heritability was observed for all fibre traits in this study. GAM was grouped in three classes, viz., lower than 10%, moderate in 10%-20%, and higher than 20% (Johnson et al. 1955). GAM of fibre length, strength, and micronaire were moderate(10%-20%). On the other hand, lower GAMs were showed for UI and elongation percentage. Heritability and GA indicated the presence of additive genetic variation among the genotypes for the traits. When high heritability is combined with high GA, researchers should adopt selection strategies in the later generations for improving these traits of crops (Rahman 2016).

Among the clustered data set, significant components were identified, which contributed to the majority of variability. In this study, higher level of variability was found in fibre strength, length, and UI. These traits distinguished all the genotypes in higher order than other traits. In Biplot analysis, variables were plotted on the vector plane and vectors were used to distinguish



**Table 6** Superior *G. barbadense* accessions for premium fiber traits

Genotypes	UHML /mm	Genotypes	MIC	Genotypes	STR / (g·tex <sup>-1</sup> )
GSB 39	40.3	ERB 4488, EA206, 25-1-3, ICB 143, CBS 148, ERB 4492, EC 959, 172, ICB 45, EC 959, 106, ICB 173	4.2	GSB 39	45.6
ERB 13, 738, CCB 33, CCB 64	39.4	26-1-3, EA197, ICB 254, ICB 196, EC 959, 188, ICB 176, 199-17-16, EC 101, 789/1, EC 959, 173, EC 959, 191, EC 134, 388, CCB 30, EC 9257, ERB 13, 750	4.1	SB. SG 1-5	43.7
CCB 143-B	39.3	ICB 29, GSB 40, EC 101, 784, ARBB 1501, ICB 172, EC 97, 639, ARBD 27, DB 1602, EC 97, 628, ICB 121, ERS 13, 758	4.0	CCB 64	43.3
SB. SG1-5, ARBB 20	39.2	5746 U, BAHTIM 185, EC 101, 783, EA196, ARBB 1402, DB 1701, DB 1402, EC 101, 793, EC 959, 184, EA203, ICB 175, SB.SG 1-5, ERB 13, 738	3.9	CCB 143-B, ICB 176	42.3
EC 959, 098	38.7	GSB 43, GSB 39, DB 1401, CV 76, ARBB 1401, EA205, ARBB 1502, GSB 45, EC 959, 098, GIZA 70, 19/2, CCB 129, ICB 104, ICB 177, EC 143, 582, GIZA 45, EC 959, 171, ARBB 20	3.8	EC 959, 191	42.1
GSB 41, ICB 176, CCB 129	38.3			ICB 254	41.9
SILS 7, EC 154, 784	37.8			CV 76, ERB 13, 738, ARBB 20	41.4
CV 76	37.3			EC 959, 189	41.3
CCB 51-2	37.0			GSB 41, MINAXI	40.7
CCB 30	36.8			CCB 33, SILS 7	40.5
EC 959, 189, GSB 45	36.3			EC 154, 784, EC 959, 171, ICB 115	40.4
EC 959, 173	36.0				
20/2 W, BAHTIM 185	35.8				
14-15 BAR	35.7				
ERB 13, 750	35.6				
ARBD 27, ICB 121, ICB 104	35.5				
EC 134, 388, EC 9261, ICB 115, ICB 177	35.4				
ICB 254	35.3				
EC 959, 191	35.2				
EC 959, 171, ARBB 1402	35.1				
Population mean	33.9		4.0		36.5
CD @5%	3.1		0.6		2.4

UHML: Upper half mean length, MIC: Micronaire, STR: Strength

variables. In our study, biplot diagram showed that fibre UI, strength, and length contributed significantly towards diversity. Micronaire and elongation percentage were short in vector length, and they had less distinguishing power. Extremely diverse genotypes, DB 16, EC 959, 191, GSB 39, ARBB 20, 5746 U, EA 203, and EA 201 were plotted on the verge of biplots. GSB 39, which having higher fibre length (40.3 mm) and strength (45.6 g·tex<sup>-1</sup>), was found to be an outlier among all genotypes. This genotype could be a potential donor for improving fibre

length and strength. Micronaire of sixty-six accessions were graded as A level (3.7-4.2).

Association among different traits is important in breeding strategies. Strong association between traits plays a vital role in direct or indirect selection of particular genotype for a given objectives (Manan et al. 2022). In our study, positive correlation was observed between fibre length, strength, and uniformity, respectively. However, these traits were negatively associated with micronaire. The present results are consistent with the findings



of previous papers (Ulloa and Meredith 2000; Karademir et al. 2011; Clement et al. 2012; Manivannan and Wagmare 2020).

Tremendous breeding efforts have been made in cotton for improving fibre length. *G. barbadense* in general has higher genetic potential for fibre length as it falls in the category of ELS. It is also a preferable donor for fibre length improvement in *G. hirsutum* backcross breeding. As fibre length has high heritability, it links with spinning performance of the yarn, spinning limit, yarn strength, yarn evenness, and product quality. Longer fibres are the source for manufacturing fine yarns (Jamshed et al. 2016). There are 34 genotypes in this study have the fibre length of more than 35 mm; these lines are useful for selection and further yield improvement. The tensile strength mainly determines the yarn strength (Chandra and Sreenivasan 2011). Based on fibre strength, advanced spinning technologies can be employed for yarn manufacturing; especially it will be preferred in modern spinning mills (Felker 2001; Zhang et al. 2019). The strength of eighteen lines are more than 40.4 g·tex<sup>-1</sup>. Along with fibre length, fibre fineness is also an important character that determines whether fibre structure is coarse or fine. Micronaire influences dyeing (Rodgers et al. 2017). Generally, micronaire values below 3.5 indicate weak structure and immature fibres that could be broken easily during spinning, and they will absorb the dyes poorly, which may create fibre entanglement also called as naps while fabric manufacturing and ultimately affect uniformity and yarn quality (Ghanmi et al. 2017). Micronaire of the 108 *G. barbadense* accessions in this study was between 3.2 and 5.9; 66 accessions were graded by micronaire values as A (best quality, 3.7 to 4.2). In *G. barbadense*, genotypes exhibiting higher levels of fineness and maturity are preferred for fine cloth manufacturing. Textile industries also prefer the micronaire value of 4.5 to 5.5 for fine and good grip cloths. Finer fibres allow a greater number of individual fibres per cross section of yarn that is an inbuilt strength of yarn, resulting in finer yarn (Han et al. 1998). However, micronaire is not an ideal index for measuring fineness (fibre maturity × linear density).

Despite our limited understanding of the genetic and physiological factors that influence fibre growth and quality, some advances in fibre quality have been made. Within current *G. barbadense* available germplasm pools, incremental improvements in fibre quality are still feasible. Nevertheless, if history is any indication, future breakthroughs in fibre quality might depend on the addition of new germplasms to the working gene pool in order to increase the genetic variability for fibre quality in *G. barbadense*.

## Conclusion

Our study revealed wide genetic variability for the fibre traits among the 108 exotic accessions of *G. barbadense*. Based on correlation studies, it was found that fibre length is strongly and positively associated with fibre strength; they are negatively correlated with micronaire, respectively, which infers that the longer fibre would be the stronger one. It provides an opportunity to use the intrinsic association present among various fibre traits for further selection programmes. PCA revealed that PC1 and PC2 accounted for 67.92% and 16.46% of the variation, respectively. It implies that all fibre traits studied contributed to discriminate the genotypes. However, UI, fibre length, and fibre strength contributed in greater magnitude towards total variation among all traits. Biplot analysis results showed the extremely diverse genotypes DB 16, EC959191, GSB 39, ARBB 20, 5746U, EA 203, and EA 201 were plotted on the verge of biplots. These could be used as parents in further crossing programmes. Screening the germplasm is helpful to identify the superior lines with fine fibre properties. Thirty-four genotypes with fibre length of more than 35 mm, 18 genotypes with fibre strength of more than 40.4 g·tex<sup>-1</sup>, and 66 genotypes with A grade (3.7 to 4.2) micronaire are identified as genotypes with superior fibre quality.

## Methods

### Germplasm accessions and field trial

A set of 108 accessions of *G. barbadense* germplasm were grown in Augmented Block Design (ABD) with three check varieties Suvin, CCB 51 and CCB 143B (Supplementary Table S1). These accessions were grown during Kharif season (July to December) of 2020–2021 at Research Farm of ICAR-CICR, RS, Coimbatore, South India (11.016 8°N, 76.955 8°E, and an altitude of 432 m above the mean sea level). Each accession were grown in a single row of 4.5 m length with checks. Checks were replicated thrice as per the ABD across ten blocks. The plants were grown with the spacing of 90 cm × 60 cm and ICAR recommendation of packages of cultivation were followed throughout the crop growing period. Random five plants were selected for each accession. Fully matured and opened bolls were harvested from selected plants, which were properly dried for fibre quality analysis. Ginning of seed cotton from the selected plants was done through cloy gin in the laboratory. After ginning, lint samples from the selected plants of each accession were bulked together to make it as homogeneity for fibre testing. Prior to fibre testing, lint was placed in an air-conditioned room with a humidifier at 65% humidity and 18–20 °C.

### Fibre quality testing

Fibre samples were prepared after ginning processes for quality testing by the High Volume Instrument (HVI-900, USTER, US) (ASTM 2005). Twenty-five gram of lint sample from each accession were taken for fibre testing. The test was repeated for three times, the mean value was recorded as final result. Fibre length in terms of upper half mean length (UHML, mm), fibre strength or tenacity ( $\text{g}\cdot\text{tex}^{-1}$ ), micronaire, fibre elongation percentage (%), and uniformity index (UI, %) were recorded for the accessions. The fibre quality testing was done at ICAR-CIR-COT, RU, Coimbatore, India.

### Statistical analysis

Analysis of variance (ANOVA) was performed to determine the genetic variation among the accessions. Variability parameters like phenotypic variance (VP), phenotypic coefficient of variation (PCV), genotypic variance (VG), genotypic coefficient of variation (GCV), heritability in the broad sense ( $H^2$ ), genetic advance (GA), genetic advance based on mean (GAM) were estimated based on Singh and Chaudhary (1985). Data were analyzed using the software STATGRAPHICS, Centurion XVI (StatPoint Technologies Inc., Warrenton, VA, USA). Correlation between these fibre traits were derived as per the Pearson's correlation coefficient using the SPSS 22.0 software. Multivariate analysis such as correlation (association analysis), principal component analysis (PCA), biplot analysis, and hierarchical clustering based on unweighted paired group method using arithmetic average (UPGMA) were done using the software PAST 4.03 (Paleontological Statistics Software Package for education and data) (Hammer et al. 2001).

### Supplementary Information

The online version contains supplementary material available at <https://doi.org/10.1186/s42397-023-00153-y>.

**Additional file 1.**

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### Authors' contributions

Alagarsamy M has done all the work single handedly.

### Funding

Not applicable.

### Availability of data and materials

The datasets used during this study can be provided on reasonable request.

### Declarations

#### Ethics approval and consent to participate

Not applicable.

#### Consent for publication

Not applicable.

#### Competing interests

The author has no conflict of interest.

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