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Research Article

Site regression and multivariate analysis for genetic diversity in *Gossypium barbadense* accessions

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Abstract
In India, *Gossypium barbadense* is cultivated in niche areas in the south and central regions of the country, especially for its quality fibre. Initially, 50 *barbadense* accessions were screened for their stability across 2 years from the germplasm maintained at ICAR, CICR, RS germplasm, and stable diverse genotypes were selected for further breeding. The results indicated that ICB 262, ICB 174 and ICB 73 were early maturing. ICB 264, CCB 26 and ICB 73 showed tolerance to sucking pests due to their higher trichome density (TD), gossypol glands and epicuticular wax (ECW). G × E interactions were analysed for the 2019–2020 and 2020–2021showed that traits like number of bolls, single plant yield and fibre strength could express better during 2019–2020, single boll weight and micronaire were dominant in season 2. Traits like ginning percentage and fibre length were stable across environments indicating that their expression is genetically controlled. PCA results indicated that about 91.45% of total variability was explained by PCA1, 2 and 3. Diversity analysis grouped the accessions into four major clusters with two sub-clusters in groups 2, 3 and 4, hence forming seven groups. The minimum distance was observed between ICB 176 and CCB 11 (0.56) and the maximum distance was observed between ICB 174 and ICB 1 (3.11).

Keywords: *Gossypium barbadense*, diversity, fibre quality, stability, variability.

INTRODUCTION

India is a country where all four spinnable fibre-yielding cotton species – *Gossypium arboreum*, *G. herbaceum* of diploid, and *G. hirsutum* and *G. barbadense*– of tetraploid cotton are grown. A wide range of variability is available in all traits: 13–37 mm span length, 25–43 g/tex tenacity, 1.4–7.4 g boll weight, etc. With such a huge variability, the right combination of length, strength and micronaire is missing in most of the popular cultivars across the world. India accounts for around 37% of the global cotton area and contributes to 24% of the production of cotton and is now the largest producer of raw cotton and the second largest consumer (cotcorp.org.in). However, Indian mills are forced to import 15% of their domestic requirement from the USA, Egypt, Sudan, etc. The import mainly includesextra-long staple (ELS)cotton (>32.5 mm SL), which is required to produce the finest yarn. The ELS cotton fibres obtained from pure *barbadense* are becoming non-remunerative due to low yield of *barbadense* and its location-specific cultivation. Hence H × B hybrids assume a special status owing to their ELS fibres. The first step in breeding for ELS cotton is to select stable pest-tolerant parents of *barbadense*.

Released in 1978 from the ICAR-Central Institute for Cotton Research, Regional Station (CICR-RS), Coimbatore, the cultivar Suvin is known to be the finest cotton produced in India. Suvin was derived from a cross between Sujatha (Indian variety) and St. Vincent (Sea Island variety) and has spinnability of up to 240s counts. The period 1989–1990 witnessed the highest production – about 36,000

bales (170 kg) of Suvin. Thereafter, the production of Suvin declined steadily over the years and now the production stands at about 1250 bales (Ministry of Textiles, 2017). When considering the Indian consumption of ELS cotton, it is likely to remain high in the years to come. At present, varieties/hybrids with the right combination of staple length range of 33–36 mm combined with micronaire of 4.0–4.5 and strength of 27–35 g/tex with better yield and wider adaptability are in great demand.

Cotton breeding has to be carried out keeping in mind two different criteria. The first criterion is the stability of the parent material across the environment. The second criterion is that the parent material should be diverse enough to obtain variability in further generations. Stability and genotype × environment interactions of genotypes could be studied by several methods for yield-related traits through conventional analysis. Different models were proposed for stability variance, eco-valence, regression coefficient analysis or principal component analysis (PCA) (Finlay and Wilkinson, 1963; Eberhart and Russell, 1966; Perkins and Jinks, 1968; Freeman and Perkins, 1971; Shukla, 1972; Kang, 1993). Kang (1993) proposed yield stability static (Ysi) by combining yield and stability as a single selection criterion by modifying the rank sum method. However, they require a minimum of three locations/year/environment data. Whereas the genotype main effect plus genotype environment interaction (GGE) also known as site regression analysis biplot graphically represents the G and GEI effect present in the multi-locational trial data using environment-centred data with two environments. GGE biplots were used to evaluate (1) mega environment analysis, where genotypes can be recommended to specific mega environments; (2) genotype evaluation, where stable specific genotypes can be recommended across all locations; and (3) location evaluation, where the discriminative power of target locations for genotypes under study is explained (Balakrishnan et al., 2016). The objectives of this study were to (1) identify stable high-yielding *G barbadense* genotypes and (2) select diverse parents for further genetic dissection and utilization in crop improvement.

MATERIALS AND METHODS

This study was conducted at the main farm of ICAR-CICR-RS, Coimbatore, during the 2019–2020 (Season I/environment I) and 2020–2021 (Season II/environment II) kharif seasons. Fifty *G. barbadense* genotypes were raised and morphological observations were recorded. The stability and diversity of the genotypes were the main parameters for inclusion in the breeding programme. **Table 1** provides a description of the germplasms used in the study. The experiment was conducted in a completely randomized block design with three replicates in both years. Five rows of each genotype were sown in a replication. Two seeds were dibbled on each hill. Eight plants were maintained per row. The spacing adopted was 90 × 60 cm. Gap filling and thinning were done after 15 days to maintain a single plant per hill. Recommended crop production and protection practices were followed to raise a healthy crop. Morphological observations were recorded at different crop intervals. Morphological observations were recorded in twenty plants in each replication for the all the parameters presented in **Table 2**. The fibre quality traits like full spinning test were done at ICAR-Central Institute for Research on Cotton Technology, Regional Station, Coimbatore and the results were obtained in HVI mode.

Epicuticular wax: About 15 ml of chloroform was taken in a test tube and known volumes of leaf bits were immersed. The tubes were shaken well for 20 seconds and the chloroform was decanted and kept over a water bath to evaporate the content. About 5 ml of potassium dichromate reagent was added and the tubes were boiled for 30 minutes. The tubes were then cooled and the volume was made up to 12 ml using distilled water. The solution was read at 590 nm absorbance and the ECW content was expressed in µg/cm^2.

Trichome density and number of gossypol glands: The TD of leaf, calyx and corolla of different *G. barbadense* accessions were estimated by following the method of Maite *et al.* (1980). Samples collected at random from plants were cut into one square centimetre size and boiled in 20 ml of water in small glass vials for 15 minutes in a hot water bath at 85°C. The water was then poured out, retaining the leaves and boiled after adding 20 ml of 96% ethyl alcohol for 29 minutes at 80°C. The alcohol was discarded and 90%lactic acid was added and stoppered

Table 1. List of germplasms used in the study

S. No.	List of germplasms	Special trait of importance
1	ICB 174, ICB 99, ICB 264, ICB 284, CCB 25, ICB 46, ICB 124, ICB 34, ICB 1	Insect tolerance/Resistance traits
2	ICB 258, CCB 11 A, ICB 183	Early maturing lines
3	ICB 39, ICB 161	Compact type
4	ICB 176, ICB 207, ICB 244, ICB 77, ICB 290, ICB 199, ICB 198, ICB 53, ICB 61, ICB 58, ICB 200	Fibre-related traits (higher GP, length, strength and fineness)
5	ICB 273, ICB 73, ICB 28, ICB 177, ICB 220, ICB 13, ICB 16, ICB 255, ICB 184	Higher number of bolls
6	CCB 141, CCB 64, ICB 40, ICB 96	Higher boll weight
7	ICB 262, ICB 35, ICB 194, ICB 75, ICB 86, CCB 143, CCB 28, CCB 11, CCB 143 B, CCB 29, ICB 129, CCB 26	Single plant yield

Table 2. Morphological observations recorded with several plants under each category

S. No.	Characters	Note
1	Date of first flowering	Early (<50 days), medium (50–60 days), late (>60 days)
2	Days to 50% flowering	Very early, early, medium, late, very late
3	Plant height	Dwarf (<60), Semi-dwarf (60–90), medium tall (91–120), tall (121–150), very tall (>150)
4	Leaf colour	Light green, green, light red, dark red
5	Leaf hairiness	Sparse, medium, dense
6	Leaf petiole pigment	Present or absent
7	Leaf appearance	Cup or flat
8	Epicuticular wax, trichome density, number of gossypol glands	Low, medium, high
9	Stem hairiness	Smooth, sparse, medium, dense
10	Stem pigment	Present or absent
11	Boll colour	Green or red
12	Boll shape	Round, ovate, elliptical
13	Boll surface	Smooth or pitted
14	Boll: prominence of tip	Blunt or pointed
15	Boll bearing	Solitary or cluster
16	Number of bolls per plant	Low, medium, many
17	Boll weight	Very small (<3.0), small (3.0–4.0), medium (4.1–5.0), large (5.1–6.0), very large (>6.0)
18	No. of monopodia per plant	Low, medium, many
19	No. of sympodia per plant	Low, medium, many
20	Seed index (g)	Very small (<5.0), small (5.0–7.0), medium (7.1–9.0), bold (9.1–11.0), very bold (>11.0)
21	Ginning outturn (%)	Very low (≤30), low (31–32), medium (33–34), high (35–36), very high (≥37)
22	Lint index (g)	Minimum, medium, maximum
23	Seed cotton yield (g/plant)	Low, medium, high
24	Lint yield/plant (g)	Low, medium, high
25	UHML (mm)	Short (≤20), medium (20.5–24.5), medium long (25.0–27.0), long (27.5–32.0), extra-long (≥32.5)
26	Uniformity index (%)	Poor (<42), fair (42–43), average (44–45), good (46–47), excellent (>47)
27	Fibre maturity ratio (%)	Very immature (≤31), immature (32–49), average (50–65), good (66–80), very good (≥81)
28	Fibre strength (g/tex)	Very weak (≤16), weak (17.0–20), medium (21.0–24.0), Strong (25.0–28.0), Very strong (≥29)
29	Fibre micronaire value	Very coarse (≥6.0), coarse (5.0–5.9), medium (4.0–4.9), fine (3.0–3.9), very fine (≤3.0)
30	Fibre strength to length ratio	Low, medium, high
31	Fibre elongation (%)	Low, medium, high

Date of first flowering (DFF), days to 50% flowering (DFPF), plant height (PH) (cm), leaf colour (LC), leaf hairiness (LH), leaf petiole pigment (LPP), leaf appearance (LA), ECW (µg/sq.cm), TD (Nos/sq. cm), number of gossypol glands per sq. cm of leaf, bracts and petals (NGG) (Nos/sq cm), stem hairiness (SH), stem pigment (SP), boll colour (BC), boll shape (BS), boll surface (BSF), boll: prominence of tip (BT), boll bearing (BB), number of bolls per plant (NB), boll weight (BW) (g/boll), number of monopodia per plant (NM), number of sympodia per plant (NS), seed index (g) (SI), ginning outturn (%) (GP), lint index (g) (LI), seed cotton yield (kg/ha) (SCY), lint yield (g/plant) (LY), upper half mean length (UHML), uniformity index (UI), fibre maturity ratio (%) (FM), fibre strength (g/tex) (FS), fibre micronaire value (µg/inch) (FF), fibre strength to length ratio (FSL) and fibre elongation (%) (FE).

and heated at 85°C for 30–45 minutes until the segment cleared.

GXE analysis: Site regression analysis, also called GGE (genotype main effect plus genotype environment interaction), was analysed using GEA-R software. The basic model is as follows:

$$Y_{ij} = \mu + e_j + \sum_{n=1}^{N} \tau_n \gamma_{in} \delta_{jn} + \varepsilon_{ij}$$

where Y_{ij} is the yield of the *i*th genotype (i = 1,… ,I) in the *j*th environment (j = 1, … ,J),μ is the grand mean,$e_j$isthe environment deviation from the grand mean,T_n is the eigenvalue of the PC analysis axis n,Y_{in} and $\delta_{jn}$are the genotype and environment principal components scores for axis n, N is the number of principal components retained in the model and ε_{ij} is the error term.

Statistical analysis: Morphological and yield attributes of the 2 years were averaged, and the pooled data were then used for analysis. First, a scoring system was created out of all the morphological data to mimic qualitative features. Using SPSS 16.0, a PCA study of the data matrix was conducted to determine the selection criteria and identify the key morphological traits that contribute significantly to variety. For further investigation, the PCs with eigenvalues >1 were chosen (Jeffers, 1967). The study of morphological characters that were not invariant or substantially linked with another character was excluded from further analysis.

Cluster analysis: DARwin was used to calculate the level of diversity present among the *G barbadense* genotypes (Perrier and Jacquemond-Collet, 2006). To uncover genetic linkages, a dissimilarity matrix for morphological observation was built using the Rogers–Tanimoto coefficient of associations. To create a dendrogram using DARwin 5.0, these data were submitted to the unweighted pair groups method with arithmetic mean (UPGMA) analysis, and dissimilarity was calculated based on the corresponding morphological scoring.

RESULTS AND DISCUSSION

Cotton being a commercial crop, the variability in the cultivated types has reached a plateau. The available germplasm collection always serves as an important tool to overcome the problem. Exploring the existing variation in the germplasm is much important to avoid the genetic vulnerability of the crop towards the newly emerging pests and diseases. Fifty *G. barbadense* genotypes were analysed for their diversity and stability for various characters in two consecutive years. About 42 morphological characters were recorded in all the genotypes. Based on DFPF, ICB 262, ICB 174, ICB 73, ICB 255, ICB 220, ICB 28, ICB 177, CCB 141 and CCB 11A were identified as early maturing lines. Breeding for selection towards a particular trait is always an advantage to cultivar improvement. Cotton is an annual crop and bringing in earliness is one of the universal objectives of the cotton breeding programme. Other advantages include decreased insect pressure, higher quality, lower costs and frequently higher yields. The variety's maturity is determined by its genetic complement, but it can be slightly altered by several edaphic factors such as soil moisture, fertility level, temperature, cloudy weather and pest pressure (Guthrie et al., 1995). Hence the number of days needed to reach physiological maturity in the identified early lines could change based on the prevailing environmental conditions.

Sucking pest resistance is comparatively low in *barbadense* genotypes than in other cotton species. The characteristics related to pest and disease resistance were studied and it was found that leaf TD was higher in ICB 124 (135/sq cm) (**Fig 1a)**.Genotype CCB 64(10 sq cm-1) had the lowest leaf trichome density, furthermore ICB 174 (125 sq cm-1) was found with the highest bract trichome density, ICB 129 (4 sq cm-1) with the lowest bract trichome density, ICB 46 (293 sq cm-1) with the highest trichome density on floral petals and ICB 200 with no floral trichome (**Fig 1 b-f)**.Genotype ICB 264 (126/sq cm) and CCB 64(6/sq cm) had higher and lower gossypol gland per sq cm of leaf area respectively (**Fig 1g,h**). Similarly, ICB 1 had higher epicuticular wax content,that is 29.63 µg/sq.cm.Genotypes ICB 264, CCB 26 and ICB 73 showed lesser insect incidence than the other entries. Correlation studies showed that most of the sucking pests had a negative correlation with number of gossypol glands in leaf and epicuticular wax content. Few cotton lines exhibited resistance towards pests due to trichome density, epicuticular wax and gossypol content. The pest population usually varies from genotype to genotype due to the external or internal physiology of the plant. Parnell *et al.* (1949) showed a negative relationship between hairiness and jassid resistance. Other morphological characteristics like hairiness, colour, thickness, toughness of tissue,and physiological (osmotic concentration of cell sap) and biochemical traits, especially gossypol content, nectar gland, tannin content, phenol compound, of the host plant are known to confer insect resistance in crop plants (Painter, 1951). This is due to the fact that plants have the ability to alter the behaviour of feeding insects (Karban and Baldwin, 1997) through the accumulation and excretion of toxic exudates, or host plants can act as barriers to insect pests due to morphological traits (Stadler, 2000; Hirota and Kato, 2001; Goncalves-Alvim *et al.,* 2004). This is due to the thick waxy cuticular layer that acts as a defence against herbivorous insects (Taiz and Zeiger, 1998). Moreover, genotypes with higher epicuticular wax show lesser disease incidence. Genotypes like CCB 64 with fewer gossypol glands can be used to generate cultivars with no gossypol glands for culinary purposes.Genotype ICB 99 had zero branching habit, ICB 176 had higher GP (37.5), FL (35.4mm),

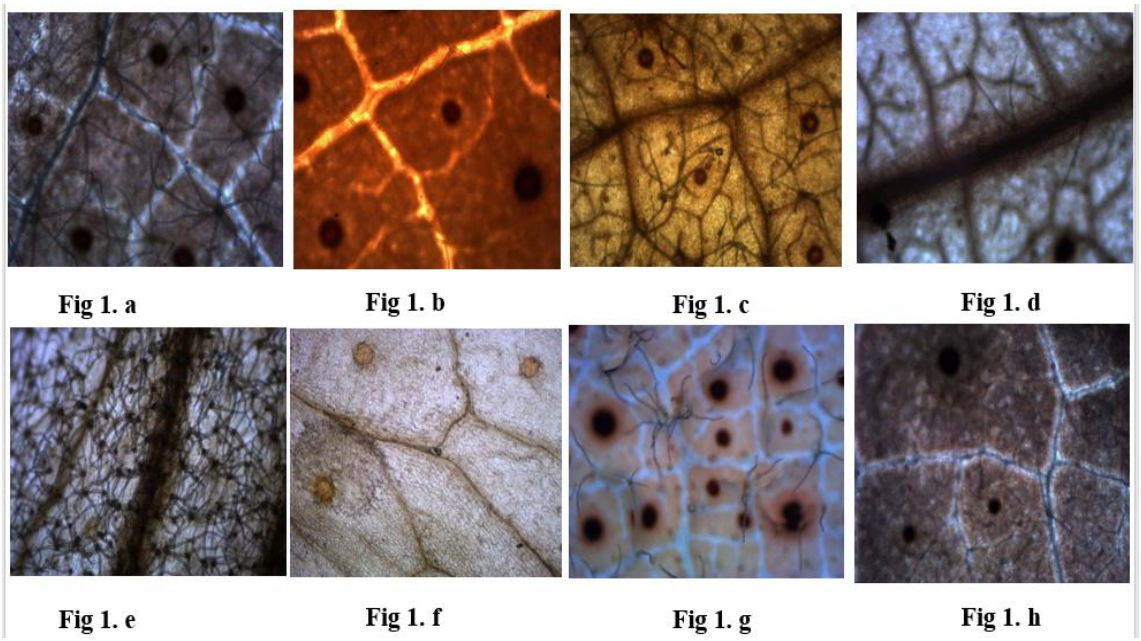

Fig. 1. Trichome and gossypol glands of leaves, bracts and flowers

(a) ICB 124 (135 sqcm^{-1}) with the highest leaf trichome density.(b) CCB 64(10 sqcm^{-1}) with the lowest leaf trichome density.(c) ICB 174 (125 sqcm^{-1}) with the highest bract trichome density.(d) ICB 129 (4 sqcm^{-1}) with the lowest bract trichome density.(e) ICB 46 (293 sqcm^{-1}) with the highest trichome density on floral petals.(f) ICB 200 with no floral trichome.(g) ICB 264 (126 sqcm^{-1}) with the highest gossypol glands.(h) CCB 64 (6 sqcm^{-1}) with the lowest gossypol glands.

FS (40.7g/tex), ICB 284 had uniformity index of 87%, ICB 258 had FF 4.1 µg/inch and CCB 143 B recorded higher boll weight of 5.35 g.

During 2019–2020, disease incidence of TSV ranged from 1.32 to 8.33 PDI, which affected only a few genotypes. Alternaria leaf spots ranged from 3.60 to 9.05 PDI and infected most of the genotypes. Grey mildew affected very few genotypes and ranged from 4.72 to 7.34 PDI. Rust emerged as a major disease at the end of the season and PDI ranged from 5.36 to 12.27 and infected most of the genotypes. However, CCB143B, ICB1, ICB75, ICB99, ICB124 and ICB264 were found to be free from all diseases. Similarly, during 2020–2021,disease incidence of TSV ranged from 1.25 to 4.35 PDI, which affected only a few genotypes. Alternaria leaf spots ranged from 3.45 to 19.66 PDI and infected most of the genotypes. Grey mildew affected very few genotypes and ranged from 2.65 to 5.40 PDI. Rust emerged as a major disease at the end of the season and PDI ranged from 3.20 to 10.10 and infected most of the genotypes. ICB16, ICB28, ICB75, ICB99, ICB124, ICB264, ICB273 and ICB290 were found to be free from all diseases.

Complex quantitative traits such as yield, with multiple contributing traits, are highly influenced by environmental interaction effects. The effect of the environment has to be analysed before selecting the genotype for any breeding programme, for which three location/environment data are required. Unlike other models, site regression analysis, also called GGE, enables to analyse two years/ two location data. It is a linear–bilinear model that takes location out of the equation and solely represents the result in terms of the influence of genotypes and the GEI. When the surroundings constitute the primary source of variation in the genotype and GEI contributions to the total variability, this is the model of choice. Additionally, unlike the AMMI model, this technique enables the detection of GEI with respect to the crossover effect brought on by significant shifts in the ranking of the genotypes amongst environments. G × E interactions were analysed in the 50 *barbadense* genotypes for the 2019–2020 (environment I) and 2020–2021 (environment II) seasons. The ANOVA table shows that there was significant variation between the two environments and the genotypes (**Table 3**). Further, the data were analysed and the results from the biplot were extracted (**Table 4**). In each biplot, the genotypes near the origin are considered to be stable under both seasons. Similarly, genotypes better suited in environment 1 are listed in **Table 4** for each trait and they were found to be near in the direction of vector 1. For this reason, their yield is bigger than the mean. Similarly, genotypes near in the direction of vector 2 are better suited for environment 2.Polygon view of the GGE biplot based on symmetrical scaling for a pattern of *G barbadense* genotypes in two environments are

Table 3. Analysis of variance for G × E interaction of cotton genotypes

Source	Degrees of freedom	Sum of squares	Mean sum of square	F value	PORCENT	PORCENAC
ENV	1	17961.19	17961.19	2567.30	6.01	6.01
GEN	49	169965.6	3468.69	495.80	56.89	62.91
ENV*GEN	49	110822.7	2261.69	323.28	37.10	100
PC1	49	197915.6	4039.09	571.61	70.49	70.49
PC2	47	82872.7	1763.25	249.53	29.51	100
Residuals	100	699.6146	6.70	NA	0	0

Table 4. List of genotypes showing stability for various traits for two environments

Traits	Stable genotypes	Season I (2019–2020)	Season II (2020–2021)
Number of bolls	ICB 13, ICB 16, ICB 35, ICB 184, ICB 220, CCB 141, ICB 255, CB 264	ICB 28, ICB 177	ICB 273, ICB 73
Single boll weight	ICB 39, ICB 46, ICB 96	CCB 141, CCB 64, ICB 161, CCB 26	ICB 35, CCB 11 A, ICB 40
Single plant yield	ICB 129, ICB 244, ICB 16, ICB 273, ICB 194	ICB 28, CCB 64	ICB 262, ICB 177, ICB 35
Ginning percentage	ICB 176, ICB 39, ICB 99	ICB 177, ICB 194	4ICB 290, CCB 143 B, CCB 141, ICB 77, ICB 28
Fibre length	ICB 262, ICB 177, ICB 194, ICB 61, CCB 29	CCB 28, CCB 11A, ICB 198, CCB 25	ICB 176, ICB 77, CCB 26, ICB 220
Fibre strength	CCB 141, ICB 262, CCB 29, ICB 40, ICB 73, ICB 258	CCB 28, CCB 25, ICB 53, ICB 143, ICB 40	ICB 174, ICB 77, ICB 198, CCB 26, CCB 11A
Micronaire	ICB 194, ICB 207, ICB 46, ICB 273, ICB 258. ICB 244, CCB 25, CCB 143 B	ICB 39, ICB 184, ICB96, ICB 177, ICB 28, ICB 75	ICB 73, ICB 199, CCB 141, ICB 99, ICB 290, ICB 161

presented in **Fig 2,** where (**Fig 2 A, C, E, G**) represents the Number of monopodial branches, single boll weight, single plant yield and ginning percentage. Similarly, **Fig.2 (B, D, F, H**) represents the Which-won-where plot number of monopodial branches, single boll weight, single plant yield and ginning outturn. The weather parameters for the two seasons are listed in **Table 5.**

The genotypes showed better performance concerning traits like number of bolls, single plant yield and fibre strength during season I. Similarly, the expression of traits like single boll weight and micronaire were remarkably better during season II. In this study, there was not much variation in weather parameters except rainfall. The expression of traits mainly depends on the rainfall pattern during the cropping season. When considering traits like ginning percentage and fibre length, both the environments were better suited and this shows that there is not much environmental influence on the two characters and their expression mainly depends on the genotype. Previous research shows that an additive form of gene action with partial dominance controls characters like plant height, number of monopodial branches, number of sympodial branches, number of bolls per plant and boll weight inheritance (Neelima *et al.,* 2004, Ahmad *et al.,* 2006, Abbas *et al.,* 2008, Kumboh *et al.,* 2008, Ali *et al.,* 2009, Zangi *et al.,* 2010 and Latif *et al.,* 2014, Surya Krishna *et al.,* 2021). However, Zia-ul-Islam *et al.* (2001), Saravanan *et al.* (2003), Kumari and Chamundeswari (2005) and Khan *et al.* (2009) reported different outcomes. The different results suggest that the expression of traits varies with the environment, in accordance with the present study. Wang *et al.* (2013) reported that fibre length changes according to genotype and environment, supporting the observation that this trait is more related to the genotype than to the environment. The square formation in September 2019 indicates that there was sufficient rainfall in season I than in season II. This might have minimized the square fall leading to a greater number of boll retention and thereby increasing single plant yield. The boll formation to maturation stage requires minimal water to get bigger bolls with quality fibre, which was seen during the second season where there was fewer rainfall compared to season I. Hence season I (2019–2020) favoured traits such as several bolls, single plant yield and fibre strength, whereas season II (2020–2021) favoured single boll weight and fibre fineness. Variation was higher for the trait plant height between two seasons. This was due to the higher amount of rainfall received during season I, which put forth more vegetative growth. The variation within the genotypes for plant height during season II was comparatively low when compared

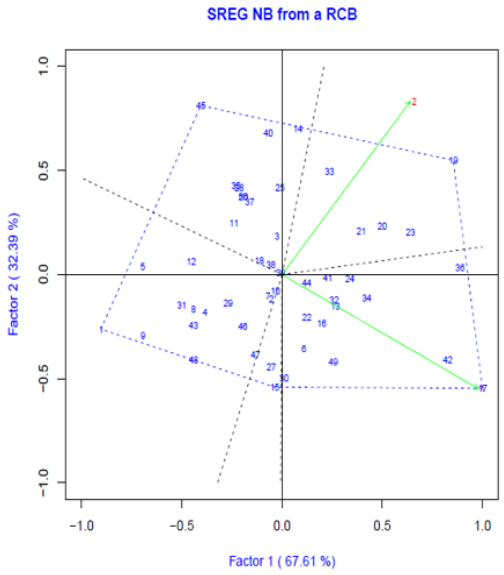

Fig. 2 A

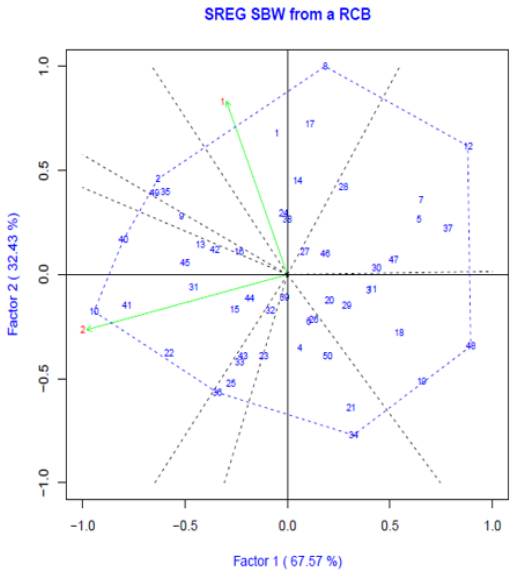

Fig. 2 C

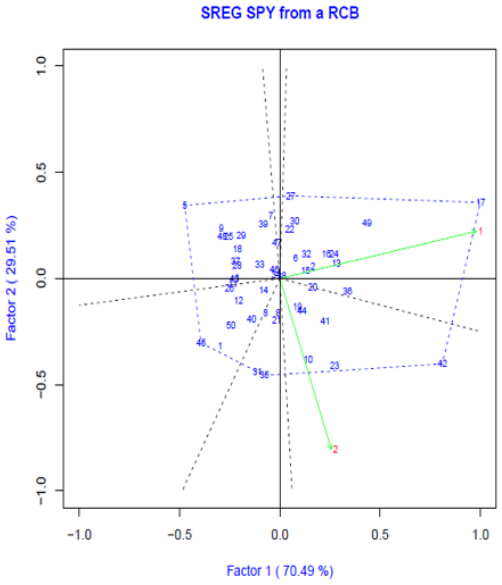

Fig. 2 E

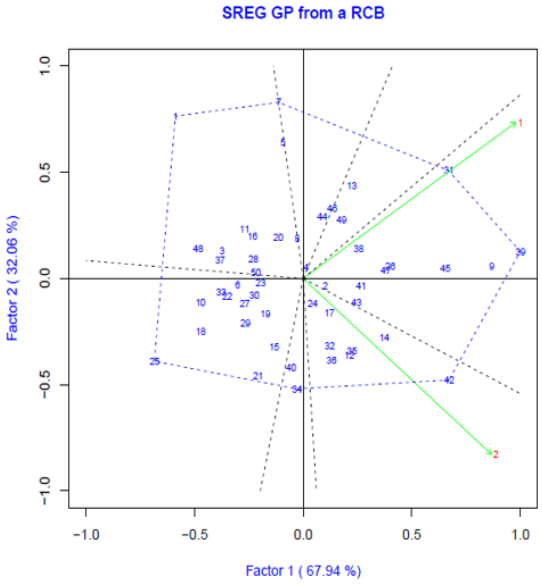

Fig. 2 G

Fig. 2. Polygon view of the GGE biplot based on symmetrical scaling for a pattern of *G. barbadense* genotypes in two environments

(A, C, E, G) Number of monopodial branches, single boll weight, single plant yield and ginning percentage

with that in season I. This shows that plant height was highly influenced by environmental variation. A medium level of environmental interactions was observed for the other yield-related characters like number of monopodia, sympodia, single boll weight, number of bolls per plant and single plant yield. The environment favoured number of monopodia, several bolls and single plant yield, as the variation was higher within the genotypes for season I. For the trait single boll weight, the variation was higher during the second season. The variation was equally distributed within the genotypes during both seasons for fibre-related traits like ginning percentage, fibre length

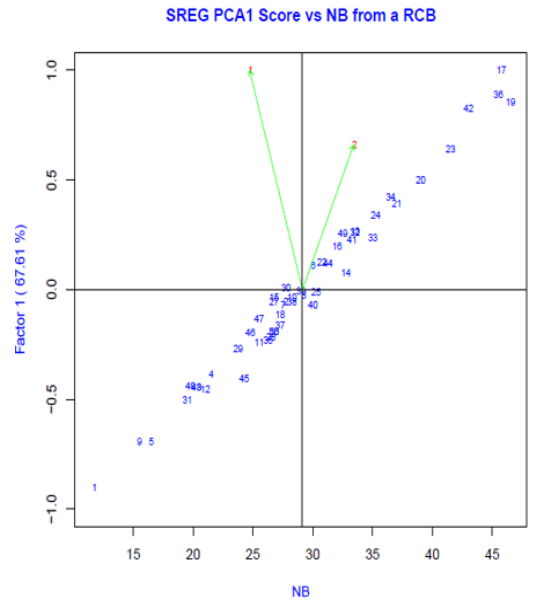

Fig. 2 B

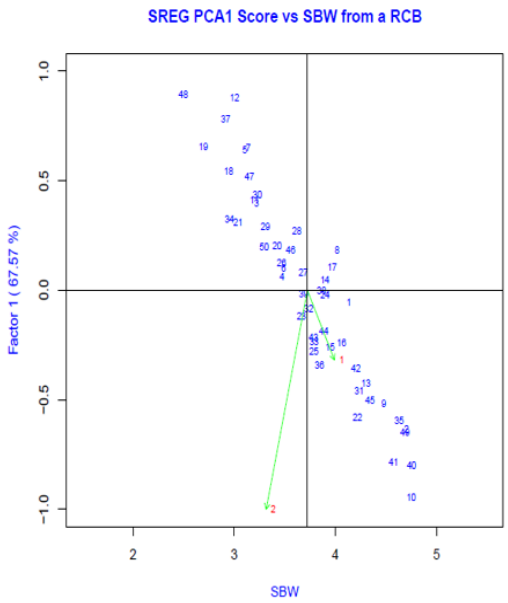

Fig. 2 D

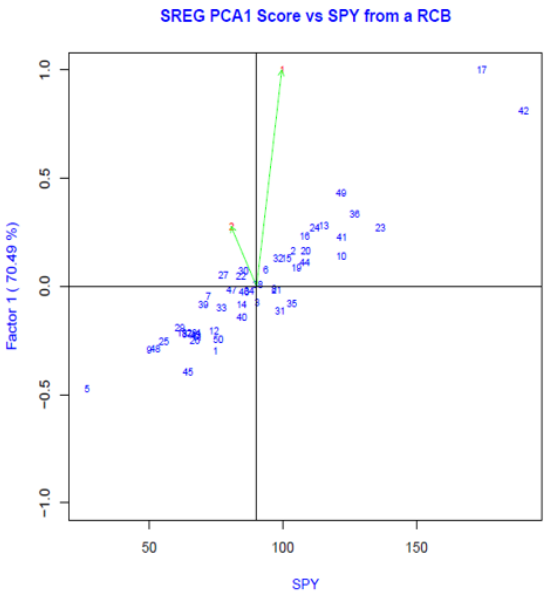

Fig. 2 F

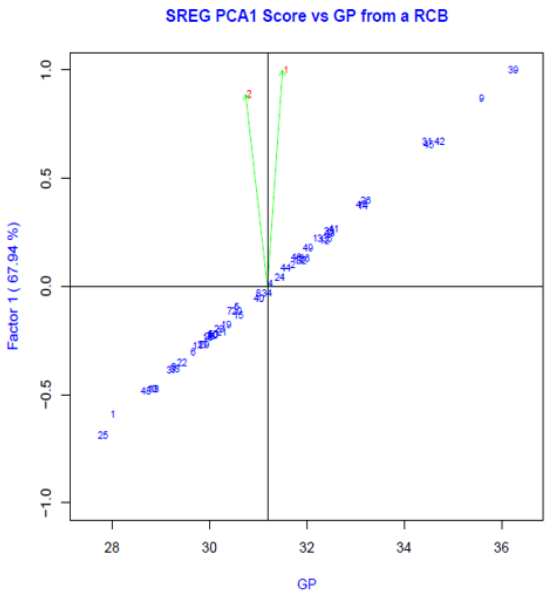

Fig. 2 H

Fig 2. Polygon view of the GGE biplot based on symmetrical scaling for a pattern of *G. barbadense* genotypes in two environments

(B, D, F, H)Which-won-where plot number of monopodial branches, single boll weight, single plant yield and ginning outturn.

(UHML) and fibre strength. The mean value was skewed for fibre fineness during season II. There were outliers for traits like number of monopodia, single plant yield, ginning percentage, fibre length and fibre fineness.

PCA measures the contribution of each variable to the total variance and also helps to estimate the impact of a particular trait on the total variance.PCA was usedto select the character harbouring maximum diversity and

Table 5. Climatic data over two seasons (*kharif* 2019–2020 and *kharif* 2020–2021)

Season	Temperature (˚C)		RH	Rainfall (mm)	Crop stage
	Max	Min			
			2019–2020		
July 19	31.8	23.7	70	8.5	Sowing
August 19	29.9	23.0	75	221.3	Vegetative stage
September 19	30.9	23.4	74.5	57.3	Square formation
October 19	30.7	22.7	74.5	246.9	Flowering
November 19	29.6	22.2	73.5	167.1	Boll formation
December 19	27.8	21.2	74	36.0	Fibre maturation
January 20	30.4	22.6	69.5	0.5	Harvesting
			2020–2021		
July 20	31.7	23.3	68.5	109.5	Sowing
August 20	29.9	23.1	71	26	Vegetative stage
September 20	31.4	23.4	74	105.5	Square formation
October 20	32.2	22.4	68	36	Flowering
November 20	33.2	22.8	65	103	Boll formation
December 20	27.7	22.1	77.5	32	Fibre maturation
January 21	27.6	21.0	74.95	141.5	Harvesting

further calculate the diversity index among the genotypes. The variation was explained by 11 components. About 33.24% of total variability was found to be explained by PCA1, PCA2 and PCA3 components. PCA1 explained 14% of the variation and was loaded mainly on fibre traits like mean length, UHML, fibre strength, boll shape and uniformity index. PCA2 elucidated 10% of variation with four main characters, *viz*. leaf and bract trichome density, leaf and bract gossypol glands (**Table 6**). Diversity analysis grouped the accessions into four major clusters with two sub-cluster groups 3 and 4, thus forming seven groups (**Fig. 3**). The minimum distance was observed between ICB 176 and CCB 11 (0.56) while the maximum distance was observed between ICB 174 and ICB 1(3.11). Based on the diversity analysis, genotypes ICB 99, ICB 176, ICB 264, ICB 284, ICB 258 and CCB 143 B were selected for the crossing programme. Cluster 3 was the smallest with nine genotypes having better fibre qualities. Mehlman *et al.* (1995) pointed out that PCA1 included the maximum information of original indexes and demonstrated no reflectionwhen compared with PCA2 and PCA3. The present study explains that the majority of variance is explained by fibre qualities. *Barbadense* germplasm acts as a resource for fibre variability in terms of length, strength, uniformity index, etc. It is evident from most of the earlier studies that yield and its components exhibited high genetic variability amongst the components in cotton (Khan *et al.,* 1999; Khan, 2003). Genetic diversity depends on the variability present within the species. The variation can be captured in different forms such as morphology, anatomy, physiological behaviour or biochemical features. Diversity is essential for plant breeders to develop new improved cultivars with desirable traits. The diversity analysis evaluated with morphological descriptors is direct, inexpensive, easy and does not require expensive technology. It gives the pattern of relatedness between genotypes based on phenotypic performance in a similar kind of environment and is reported to be equally effective as that of molecular markers in crops. During phenotype-based cluster analysis, Manhattan distance grouped the studied 50 *barbadense* genotypes into four major clusters with three sub-clusters. Significant variability for morphological traits in *Gossypium* accessions maintained at the Active Germplasm Bank of Embrapa was observed by Vidal Neto *et al.* (2008).

The study allowed classifying individuals into well-defined groups based on the fibre trait and leaf morphology of which the former is of economic importance and the latter is concerned with pest and disease resistance. Many studies have shown utility, demonstrating associations of single inheritance (discrete variables) with disease and pest resistance behaviour and fibre characteristics (Juhasz *et al.,* 2013). The variability thus hidden in the germplasm has high potential use in breeding programmes. The selection of parental material is a very crucial and essential step in plant breeding. The parents need to be tested across locations and years/seasons to evaluate its stability. A minimum of three years becomes mandatory to run any analysis. The programmes in which time is a major part of constraining resource, through GGE interaction, the parent's stability across location can be estimated in two years of time. Thus, this method aids breeders in plant breeding programme for parental selection. In this study, the genotype selected through GGE analysis, even though they were not high yielders, but they proved to have stable yields even during the adverse conditions.

Table 6.Total variance in *Gossypium barbadense* genotypes explained through principal component analysis

Component	Total	% of Variance	Cumulative %	Component	Total	% of Variance	Cumulative %
1	4.316	13.924	13.924	22	0.300	0.967	96.451
2	3.228	10.414	24.338	23	0.247	0.797	97.247
3	2.759	8.899	33.237	24	0.212	0.685	97.932
4	2.468	7.961	41.198	25	0.186	0.599	98.532
5	2.239	7.223	48.421	26	0.144	0.465	98.997
6	1.781	5.746	54.167	27	0.105	0.337	99.334
7	1.655	5.338	59.505	28	0.089	0.288	99.622
8	1.590	5.130	64.636	29	0.056	0.180	99.802
9	1.324	4.270	68.905	30	0.038	0.121	99.923
10	1.111	3.582	72.488	31	0.024	0.077	100.000
11	1.053	3.397	75.884	32	4.316	13.924	13.924
12	0.921	2.971	78.855	33	3.228	10.414	24.338
13	0.869	2.803	81.657	34	2.759	8.899	33.237
14	0.821	2.648	84.305	35	2.468	7.961	41.198
15	0.681	2.197	86.502	36	2.239	7.223	48.421
16	0.631	2.035	88.537	37	1.781	5.746	54.167
17	0.531	1.714	90.251	38	1.655	5.338	59.505
18	0.521	1.682	91.933	39	1.590	5.130	64.636
19	0.443	1.431	93.364	40	1.324	4.270	68.905
20	0.338	1.091	94.455	41	1.111	3.582	72.488
21	0.319	1.029	95.484	42	1.053	3.397	75.884

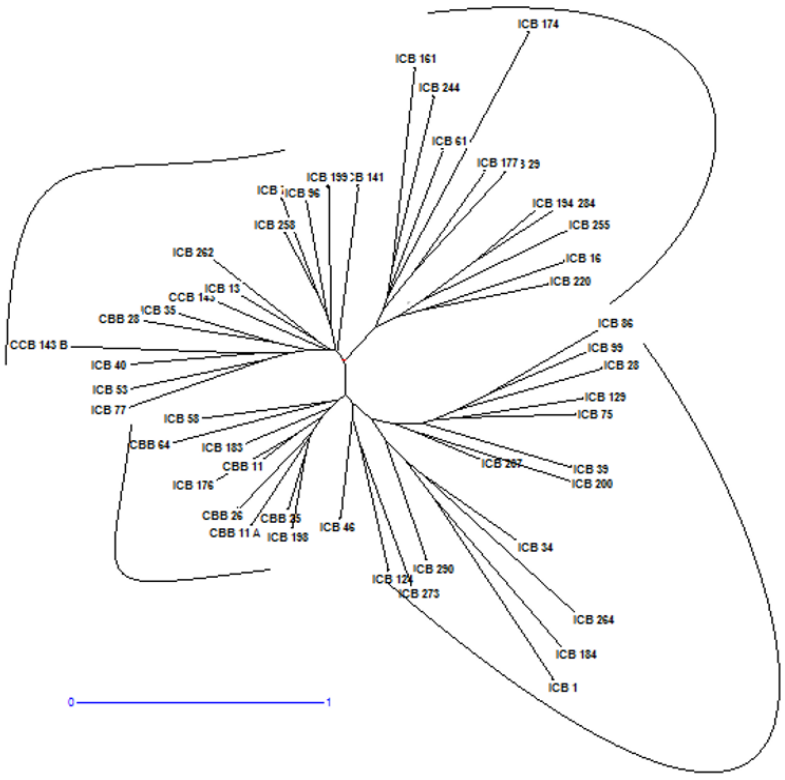

Fig. 3. Dendrogram of Gossypium barbadense genotypes constructed with four main clusters and three sub-clusters based on the UPGMA method

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