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VARIABILITY IN LEAF MORPHOLOGICAL AND SIZE CHARACTERS AMONG *CAESALPINIA CRISTA* L. ACCESSIONS FROM SOUTH GUJARAT INDIA

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ABSTRACT

Caesalpinia crista L. is an important medicinal plant widely used in traditional medicinal systems. Fifty *C. crista* accessions were investigated, collected from different locations of South Gujarat, for variability in leaf morphological and size characters. The experimental design adopted for fifty accessions was descriptive statistics for individual leaf parameters. All accessions exhibited bipinnate compound leaves with uniform qualitative traits such as leaf shape, apex, base, colour and texture. However, significant variability was observed in quantitative leaf size characters including leaf length, leaf width, petiole length, rachis length, base diameter and leaf area. Principal component analysis revealed that major variation was contributed by leaf length, leaf width and leaf area, while hierarchical cluster analysis grouped the accessions into distinct clusters based on leaf size traits. The observed variability indicates substantial genetic diversity among *C. crista* accessions, which can be effectively utilized in selection, conservation and improvement programmes.

Keywords : *Caesalpinia crista* L., medicinal plant, accessions, leaf morphology, variability

Introduction

Caesalpinia crista L. is a woody, perennial, leguminous climber or shrub with spines, widely distributed throughout tropical and subtropical regions of Asia, including China, Malaysia, Sri Lanka and India (Chan *et al.*, 2018). It belongs to the Fabaceae family and subfamily Caesalpinioideae. The species is characterized by large, bipinnate leaves bearing two to six pairs of primary leaflets (pinnae), each consisting of four to nine pairs of smaller leaflets (pinnules). Owing to its ecological resilience, adaptability to harsh environments and wide distribution, *C. crista* represents an important component of coastal and semi-arid vegetation.

The plant is an important medicinal species widely used in traditional systems of medicine. Different parts such as leaves, seeds and roots are reported to possess anthelmintic, anti-inflammation, analgesic activity, antioxidant, anti-tumour and anti-malarial activity (Upadhyay *et al.*, 2019). The therapeutic potential of *C. crista* is attributed to its rich and diverse phytochemical composition, like alkaloids, flavonoids, proteins, saponins, triterpenoid, tannins, coumarins, carbohydrates, reducing sugars, triterpenoids and fatty acids making it an important medicinal resource (Upadhyay *et al.*, 2019; Sodhi *et al.*, 2023). Proper characterization of this plant is therefore essential for its effective utilization and conservation.

Morphological characterization is a fundamental approach for plant identification, taxonomic classification and assessment of genetic variability. Among various morphological traits, leaf characters are considered highly informative due to their relative stability, ease of measurement and strong association with physiological processes such as photosynthesis and growth. Leaf macro- and micromorphological traits (leaf shape, length, width, petiole length, leaf area, venation and epidermal features) often exhibit high interspecific distinctness and often low intraspecific variability, making them reliable taxonomic markers (Qiu *et al.* 2023; Fan *et al.* 2025). Such traits are also valuable in selection and improvement programmes for identifying superior plant accessions.

Despite the medicinal and ecological importance of *C. crista*, systematic information on variability in leaf morphological and size characters among its natural populations is limited, particularly for accessions growing in South Gujarat. Detailed evaluation of leaf morphological variability is essential for effective germplasm characterization, identification of superior accessions and formulation of appropriate conservation strategies. Therefore, the present investigation was undertaken to study the variability in leaf morphological and size characters among *Caesalpinia crista* L. accessions collected from South Gujarat, with the objective of generating baseline

information useful for tree improvement, conservation and sustainable utilization of this valuable medicinal species

Materials and Methods

Study area

The present investigation was carried out in South Gujarat, India, during 2022–23 to 2024–25, which lies between 21°14'–22°49' N latitude and 72°22'–74°15' E longitude. The region falls under AES-III (Heavy Rainfall Zone) and is characterized by a sub-humid climate with temperature ranging from 6 to 45 °C and annual rainfall varying from 250 mm in the north-western part to more than 1500 mm in southern parts.

Selection of accessions

Fifty accessions of *Caesalpinia crista* L. were selected from natural populations across different districts of South Gujarat, including Navsari, Valsad, Surat, Narmada and Dang. A selective sampling strategy was used which means that only those plants showing healthy growth and desirable features were chosen, through extensive field surveys conducted during April to June 2023. Each selected plant was assigned a unique accession number and its geographical location was recorded using a Global Positioning System (GPS) (Table 1).

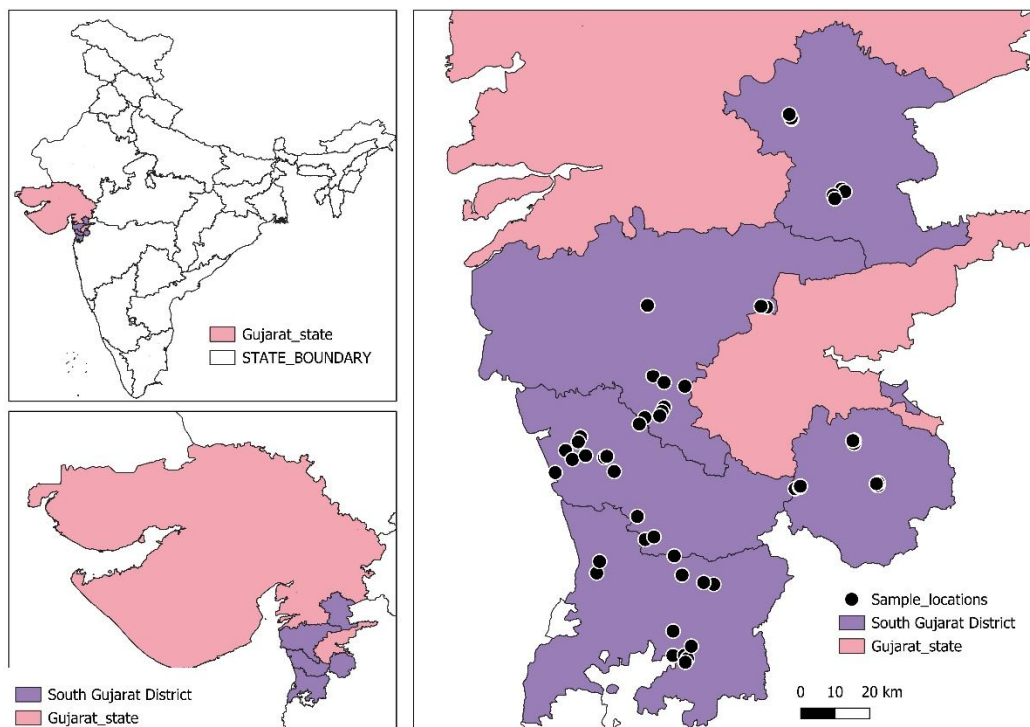


Fig. 1: Location map of selected *C. crista* L. accessions

Leaf morphological and size characters

Qualitative leaf characters such as leaf type, leaflet shape, apex, base, margin, texture and colour of adaxial and abaxial surfaces were recorded following standard botanical descriptors (Bendre and Pandey, 1996). Quantitative leaf size characters including leaf length, leaf width, petiole length, rachis length and base diameter were measured using a standard scale and digital vernier caliper. Leaf area (cm²) was measured using Biovis PSM Leaf v4.60 computer software, which enabled precise estimation of leaf surface area and captured variation among accessions.

Statistical analysis

The data recorded for leaf morphological and size characters were subjected to descriptive statistical analysis. Multivariate statistical analyses, including Principal Component Analysis (PCA) and Hierarchical Cluster Analysis (Ward's method), were performed to assess the extent of variability and grouping of *C. crista* accessions based on leaf size characters. Statistical analyses were carried out using JMP 9.0 software.

Results and Discussion

Leaf Morphological Variation

Significant variation was observed in leaf morphological traits among the fifty accessions of *C. crista*, indicating intraspecific morphological and genetic diversity (Table 1).

The leaf shape of *C. crista* accessions exhibited noticeable variation, reflecting genetic diversity and adaptation to micro-environmental conditions. Variations were observed in terms of leaf symmetry, apex and base. Leaf length varied widely, ranging from 46.00 cm (CR 05) to 78.60 cm (CR 22). The maximum leaf length (78.60 cm) was recorded in CR 22, followed by CR 25 (73.73 cm), CR 21 (73.20 cm), CR 24 (72.13 cm) and CR 31 (72.00 cm). The minimum leaf length (46.00 cm) was recorded in CR 05, followed by CR 43 (53.33 cm), CR 38 (55.33 cm), CR 34 (55.80 cm) and CR 04 (52.80 cm). Leaf width ranged from 27.40 cm (CR 43) to 44.40 cm (CR 08). The widest leaves (44.40 cm) were observed in CR 08, followed by CR 22 (44.27 cm), CR 25 (41.60 cm), CR 45 (41.40 cm) and CR 21 (41.07 cm). The narrowest leaves (27.40 cm) were found in CR 43, followed by CR 19 (29.67 cm), CR 06 (30.47 cm), CR 41 (30.47 cm) and CR 07 (30.60 cm).

Petiole length varied from 11.40 cm (CR 26) to 16.80 cm (CR 21). The longest petiole (16.80 cm) was recorded in CR 21, followed by CR 25 (15.00 cm), CR 22 (15.80 cm), CR 32 (15.80 cm) and CR 42 (15.20

cm), whereas CR 26, CR 12, CR 34, CR 30 and CR 09 recorded shorter petioles. Rachis length ranged from 33.40 cm (CR 05) to 62.67 cm (CR 22). The maximum rachis length (62.67 cm) was recorded in CR 22, followed by CR 24 (58.60 cm), CR 15 (58.20 cm), CR 20 (57.07 cm) and CR 16 (57.40 cm). The minimum rachis length (33.40 cm) was recorded in CR 05, followed by CR 43 (41.00 cm), CR 38 (43.00 cm), CR 34 (43.27 cm) and CR 04 (39.20 cm).

Leaf Base diameter ranged from 7.66 mm (CR 10) to 11.08 mm (CR 45). The maximum base diameter (11.08 mm) was observed in CR 45, followed by CR 49 (10.77 mm), CR 35 (10.73 mm), CR 50 (10.73 mm) and CR 47 (10.26 mm). The minimum base diameter (7.66 mm) was recorded in CR 10, followed by CR 01 (7.70 mm), CR 43 (7.70 mm), CR 38 (7.74 mm) and CR 30 (7.81 mm). Total Leaf Area (sq.cm) varied widely from 196.71 sq. cm (CR 42) to 1135.35 sq. cm (CR 50). The maximum leaf area (1135.35 sq. cm) was recorded in CR 50, followed by CR 48 (1097.03 sq. cm), CR 47 (1070.89 sq. cm), CR 49 (1058.42 sq. cm) and CR 02 (1064.55 sq. cm). The minimum leaf area (196.71 sq. cm) was recorded in CR 42, followed by CR 43 (197.89 sq. cm), CR 40 (223.29 sq. cm), CR 38 (229.15 sq. cm) and CR 33 (228.42 sq. cm).

Accessions such as CR21, CR22 and CR25 with longer petioles may contribute to better light interception and photosynthetic efficiency, making them promising for selection in future improvement programs. Overall, accessions CR 22, CR 45 and CR 50 exhibited superior leaf traits, while CR 05, CR 43 and CR 42 represented the lower range for these parameters, demonstrating a broad phenotypic range within the species.

Wide ranges in leaf length and width, together with large differences in total leaf area, are consistent with reports in other trees and medicinal species where leaf dimensions show strong genetic control and high heritability, making them reliable indicators of genotypic divergence and useful selection criteria in breeding and domestication programmes (Dubey and Upadhyay, 2020). Similar high intraspecific variation and strong genetic control of leaf traits have been reported in medicinal plants such as *Gymnema sylvestre* (Raghavendra *et al.*, 2024; Rohini *et al.*, 2025) and *Morinda officinalis* (Luo *et al.* 2022), where leaf dimensions were associated with genetic diversity and bioactive compound content. The marked variation in leaf length and width observed in *C. crista* suggests that these traits are largely genetically governed and may directly influence photosynthetic efficiency and biomass production. Comparable findings have been reported in *Withania somnifera* (Kumar *et al.*, 2007)

and *Withania coagulans* (Khadivi *et al.*, 2025), where extensive polymorphism in morphological traits, particularly leaf dimensions, resulted in clear clustering of accessions and reflected substantial genetic divergence.

Variation in petiole and rachis length among accessions suggests differences in canopy architecture and leaf display, traits known to influence light interception and photosynthetic efficiency in crop and tree species (Sodhi *et al.*, 2017). Accessions with longer petioles and rachises (CR 21, CR 22, CR 25) are therefore likely to possess more favourable canopy structures for light capture and biomass production. Comparable associations between stem or leaf support traits and plant vigor have been reported in medicinal trees such as *Eucommia ulmoides* (Wang *et al.*, 2023) and *Cinnamomum cassia* (Han *et al.*, 2024), where leaf size and related traits were strongly linked to growth performance, climatic gradients and, in some cases, secondary metabolite content. Such findings reinforce the relevance of base diameter as an indicator of robustness and adaptive potential. Evidence from long-lived medicinal trees further supports the genetic basis of leaf morphological diversity. In *Ginkgo biloba* (Zhou *et al.*, 2022), extensive variation in leaf area and weight among ancient populations was shown to have a clear genetic basis through genome-wide association studies, while community-level studies across forest medicinal flora have demonstrated that leaf morphological traits reflect contrasting adaptive strategies among trees, shrubs and climbers.

Table 1: Variability in leaf morphological characters among of *C. crista* accessions

Accessions Number	Leaf length (cm)	Leaf width (cm)	Petiole length (cm)	Rachis length (cm)	Base diameter (mm)	Leaf area (sq.cm)
CR 01	64.80	38.47	13.20	52.00	7.70	574.64
CR 02	66.13	35.80	12.60	53.67	8.09	1064.55
CR 03	58.87	37.80	12.40	47.47	8.31	540.70
CR 04	52.80	37.40	13.40	39.20	8.33	664.91
CR 05	46.00	34.60	13.20	33.40	9.49	502.06
CR 06	55.07	30.47	14.20	39.07	8.88	529.50
CR 07	55.47	30.60	13.60	40.80	8.73	504.13
CR 08	68.73	44.40	14.00	54.00	7.84	502.09
CR 09	60.80	34.80	13.40	47.87	8.34	496.03
CR 10	59.33	33.80	13.00	46.47	7.66	549.02
CR 11	57.20	35.07	12.60	44.40	8.06	493.01
CR 12	60.87	37.20	11.60	48.53	8.09	523.66
CR 13	66.07	40.80	14.00	51.87	8.90	729.64
CR 14	66.47	37.47	15.40	52.93	8.26	502.89
CR 15	69.80	37.00	13.00	58.20	8.40	475.47
CR 16	70.00	37.73	13.80	57.40	8.53	455.67
CR 17	64.60	38.33	12.60	52.33	8.04	429.95
CR 18	60.07	31.33	13.40	46.33	8.02	446.44
CR 19	59.60	29.67	12.80	46.47	8.19	394.93

CR 20	71.07	35.80	14.20	57.07	8.56	299.49
CR 21	73.20	41.07	16.80	54.33	8.68	459.66
CR 22	78.60	44.27	15.80	62.67	10.36	312.01
CR 23	62.33	34.33	14.00	49.40	8.18	398.22
CR 24	72.13	40.40	14.80	58.60	8.46	335.02
CR 25	73.73	41.60	15.00	56.93	8.38	267.55
CR 26	56.47	34.87	11.40	45.53	7.87	442.75
CR 27	63.93	36.13	12.80	51.27	8.66	300.81
CR 28	63.60	36.47	12.00	51.87	8.63	463.02
CR 29	59.13	34.73	12.40	46.93	8.06	256.21
CR 30	58.73	32.87	12.40	45.87	7.81	242.37
CR 31	72.00	34.00	14.00	56.93	9.11	259.59
CR 32	71.27	36.47	15.80	55.93	9.17	243.28
CR 33	62.67	36.47	14.00	49.87	9.27	228.42
CR 34	55.80	32.13	11.60	43.27	8.45	212.66
CR 35	70.80	38.93	14.80	55.07	10.73	259.21
CR 36	65.07	31.13	13.40	52.67	9.66	266.48
CR 37	66.73	32.53	13.00	52.07	9.18	232.20
CR 38	55.33	31.07	12.00	43.00	7.74	229.15
CR 39	71.20	38.33	14.20	57.60	8.96	291.03
CR 40	67.60	34.73	14.40	52.60	8.44	223.29
CR 41	61.40	30.47	10.60	50.27	7.81	251.16
CR 42	64.80	37.33	15.20	51.00	9.45	196.71
CR 43	53.33	27.40	13.00	41.00	7.70	197.89
CR 44	69.20	36.60	14.40	54.33	9.37	214.00
CR 45	71.33	41.40	15.60	54.73	11.08	991.70
CR 46	65.53	35.47	14.80	49.60	10.10	688.99
CR 47	62.60	32.80	14.20	47.67	10.26	1070.89
CR 48	60.13	33.60	12.80	46.60	9.03	1097.03
CR 49	71.67	39.20	16.00	56.13	10.77	1058.42
CR 50	69.93	35.67	15.40	53.07	10.73	1135.35
SE(m)	0.75	0.478	0.001	0.36	0.039	6.55
C.D.	2.107	1.344	0.002	1.013	0.11	18.39
SE (d)	1.06	0.676	0.001	0.51	0.055	9.26
C.V. (%)	2.027	2.313	0.011	1.24	0.771	2.41

Principal Component Analysis (PCA)

Principal component analysis was carried out to assess the variability in leaf morphological traits among fifty accessions of *Caesalpinia crista* L. The analysis reduced the six original leaf characters into a smaller number of uncorrelated principal components. The first five principal components explained 99.83 % of the total variation.

The first principal component (PC1) accounted for 54.80 % of the total variance (eigenvalue = 3.36) and showed high positive loadings for leaf length, leaf width, petiole length and rachis length, indicating that PC1 represents overall leaf size and structural dimensions. PC2 explained 21.41 % of the total variation (eigenvalue = 1.31) and was strongly influenced by leaf area and base diameter, suggesting that this component reflects variation related to leaf surface area and thickness. PC3 contributed 11.53 % of the variance and showed notable loadings for leaf area and leaf width.

Together, PC1 and PC2 together accounted for 76.22 % of the total variability, suggesting that most of the variation among accessions can be effectively explained by these two components. The scree plot showed a distinct elbow after PC2, confirming the significance of the first two components (Fig. 2).

The scree plot based on PC1 and PC2 revealed wide dispersion of accessions across all quadrants, indicating substantial morphological diversity. Separation along PC1 was largely driven by leaf size traits such as leaf length, leaf width and rachis length, while variation along PC2 was associated with leaf area and base diameter. The absence of complete overlap among accessions suggests distinct morphological differentiation (Fig. 3).

The PCA biplot further confirmed that leaf length, leaf width, petiole length and rachis length contributed strongly to PC1, whereas leaf area and base diameter were more closely associated with PC2. The close alignment of vectors for leaf length, width and rachis length indicates positive correlations among these traits, whereas differences in vector orientation suggest differential contributions to accession divergence.

Table 2: Principal component analysis of the leaf characters variability

Parameter	PC1	PC2	PC3	PC4	PC5
Leaf length (LL)	0.513	-0.241	0.021	-0.328	-0.148
Leaf width (LW)	0.426	-0.115	0.488	0.619	0.429
Petiole length (PL)	0.445	0.179	-0.390	0.464	-0.615
Rachis length (RL)	0.473	-0.344	0.131	-0.462	-0.024
Base diameter (BD)	0.347	0.499	-0.500	-0.163	0.595
Leaf area (LA)	0.113	0.727	0.585	-0.231	-0.249
Eigenvalue	3.36	1.31	0.71	0.45	0.29
Percent (%)	54.80	21.41	11.53	7.39	4.70
Cum percent (%)	54.80	76.22	87.75	95.13	99.83

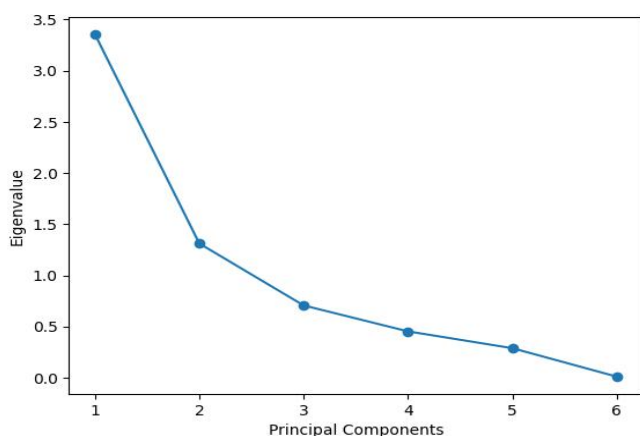


Fig. 2: Scree plot based on various leaf size characters among *C. crista* L. accessions

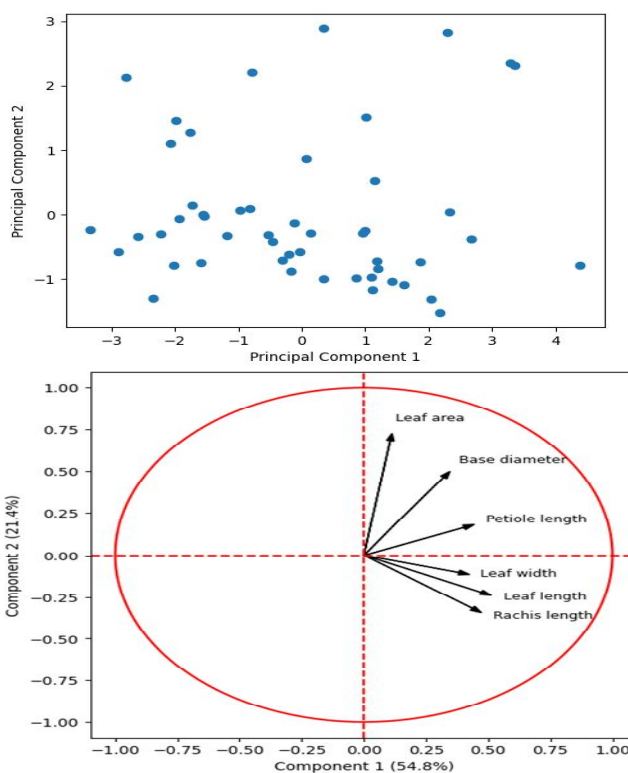


Fig. 3: Scatter plot of fifty accessions based on two principal components for different leaf size characters among *C. crista* L. accessions

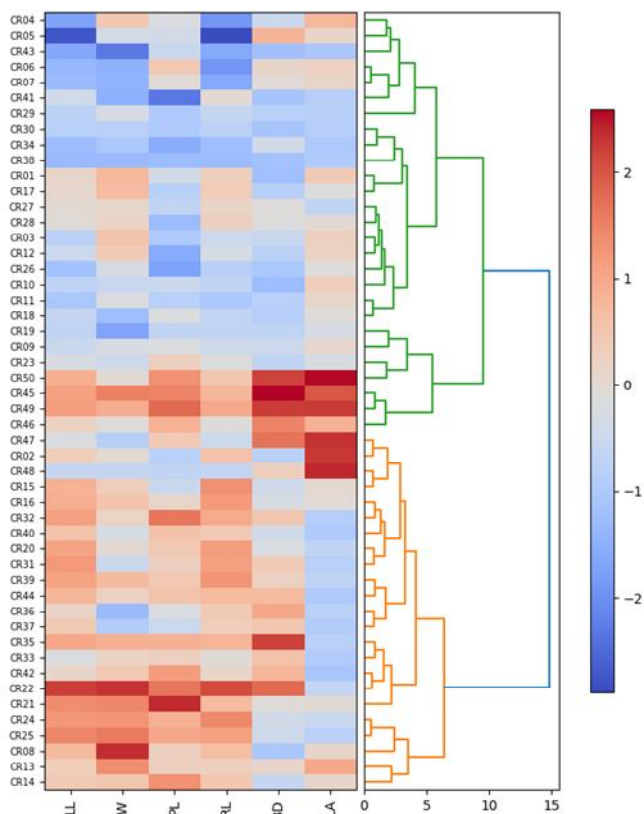


Fig. 3: Hierarchical cluster analysis (Ward method) based on leaf characters of fifty accessions of *C. crista* L.

The cluster mean analysis grouped the fifty accessions of *C. crista* L. into three distinct clusters based on leaf size characters, indicating substantial morphological variability among the accessions, confirming the presence of substantial phenotypic diversity within the species.

Cluster I, comprising 21 accessions, exhibited comparatively lower mean values for leaf length, petiole length and rachis length, indicating relatively smaller leaf size and moderate leaf area. Cluster II, consisting of 9 accessions, recorded the highest mean leaf area (568.86 sq. cm) and base diameter (9.40 mm), suggesting accessions with broader and thicker leaves. Cluster III, included 20 accessions, which showed the

maximum mean values for leaf length (70.48 cm), leaf width (38.66 cm), petiole length (14.83 cm) and rachis length (55.57 cm). The clustering pattern closely aligned with the results of principal component analysis, where these traits showed strong loadings on the first two principal components. This concordance among multivariate analyses strengthens the reliability of the observed grouping and highlights the importance of leaf morphological traits in characterizing genetic diversity.

Overall, the differences among cluster means provides valuable insight into the structure of leaf morphological variability in *C. crista* and facilitates identification of distinct accession groups.

Table 3: Cluster means leaf size characters among *C. crista* L. accessions

Cluster	No. of accessions	Leaf length (cm)	Leaf width (cm)	Petiole length (cm)	Rachis length (cm)	Base diameter (mm)	Leaf area (sq. cm)
Cluster I	21	59.77	34.29	12.53	47.29	8.08	436.77
Cluster II	9	59.92	33.07	13.69	45.75	9.40	568.86
Cluster III	20	70.48	38.66	14.83	55.57	9.21	460.60

Conclusion

The present study revealed substantial variability in leaf morphological and size characters among fifty accessions of *Caesalpinia crista* L. collected from South Gujarat. Leaf length, rachis length and total leaf area were the major contributors to variability, as confirmed by PCA and cluster analyses. Accessions CR 22, CR 45 and CR 50 showed superior leaf traits and may be prioritized for selection and conservation.

The combined application of univariate and multivariate analyses effectively characterized the extent of variability among *C. crista* accessions. The observed diversity highlights the potential of these accessions for selection, conservation and future improvement programmes. The findings provide valuable baseline information for germplasm evaluation and sustainable utilization of this important medicinal species.

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