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Kartheka T
Department of Floriculture and Landscape Architecture, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India

K Rajamani
Department of Floriculture and Landscape Architecture, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India

M Ganga
Department of Floriculture and Landscape Architecture, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India

N Manikanda Boopathi
Department of Plant Biotechnology, Centre for Plant Molecular Biology and Biotechnology, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India

Corresponding Author:
Kartheka T
Department of Floriculture and Landscape Architecture, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore, Tamil Nadu, India

Morphological characterization of certain *Jasminum sambac* genotypes using principal component analysis

Kartheka T, K Rajamani, M Ganga and N Manikanda Boopathi

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Abstract
Jasminum sambac is one of the most important traditional flower crops of India especially Tamil Nadu. Understanding the genetic variability among the different genotypes of *Jasminum sambac* helps in its exploitation in breeding programmes. A total of 34 genotypes comprising of traditional landraces collected from all over Tamil Nadu were evaluated for 40 qualitative traits according to the DUS descriptors specified for *Jasminum sambac* (Kannan M. *et al*., Plant Variety Journal, 2016). In order to reduce the dimensionality of the data, examine the variation and the relative contribution of the traits for the total variability, Principal Component Analysis (PCA) method was adopted. PCA test in 34 genotypes for all the 40 characters resulted in fifteen Principal Components (PCs) with an eigenvalue more than one accounted for 97.21 per cent of the total variability and revealed that the traits leaf margin undulation, flower bud shape, flower shape, shape of corolla lobe, flower petal tip, leaf blade undulations, flower bud length and root suckers exhibited maximum variation. Agglomerative Hierarchical Clustering and PCA results showed that the genotypes Acc. Js- 11, Acc. Js- 12, Acc. Js- 13, Acc. Js- 14, Acc. Js- 20, Acc. Js- 25, Acc. Js- 27 and Acc. Js- 32 were found to be the most diverse genotypes.

Keywords: *Jasminum sambac*, genotypes, DUS, variability, PCA, correlation, clustering

Introduction
Jasminum sambac (L.) Aiton belongs to the olive family Oleaceae. The word jasmine comes from the Arabic name 'Yasmine' meaning fragrance. *J. sambac* is a small to medium sized shrub growing up to a height of 3 m. It is one of the important traditional flower crops mainly cultivated for its fragrant flowers and highly valued for their ornamental, edible and medicinal values. India is one of the evolution centers for the genus *Jasminum* with greater diversity and hence the Tamil Nadu (Veluswamy *et al.*, 1975) [9]. Ramanathapuram gundumalli is the important *J. sambac* ecotype cultivated in Tamil Nadu. 'Madurai Malli' has been given Geographical Indication (GI) tag for its unique fragrance, exclusive shape and size. *J. sambac* phenotypes are generally classified based on the number of petal whorls present *viz.*, single-whorled, double whorled and multi whorled types. Single whorled and double whorled cultivars are the commonly cultivated types. The single whorled cultivars are highly fragrant and utilized in the perfume industry while, the double whorled cultivars yield the highest number of flowers. The traditional method for the identification of *Jasminum* species is by analyzing the morphological and physiological characters (Raman, 1955; Bhatnagar, 1956; Mohammad *et al.*, 1970) [10, 12]. Morphological characterization is essential for the identification and registration of the cultivated varieties. Hence, the present study was carried out to assess the diversity among the *J. sambac* genotypes through morphological characterization.

Materials and Methods
A germplasm comprising of 34 genotypes of *Jasminum sambac* (Table 1) collected from all over Tamil Nadu has been planted and maintained in the Botanical Garden, Department of Floriculture and Landscape Architecture, Horticultural College and Research Institute, Tamil Nadu Agricultural University, Coimbatore, India. Qualitative data on the forty characters were specified by DUS guidelines proposed by PPV & FRA (Kannan M. *et al*., Plant Variety Journal, 2016) [1] was recorded at the appropriate developmental stages.
All the observations on the leaf characteristics were recorded on the fourth fully opened leaf from the tip of the stem. Colours of the vegetative parts were observed on the plants exposed to natural growing conditions. All the colour characteristics were assessed using the latest

Royal Horticultural Society (RHS) colour charts. For assessing the stability and distinctiveness, all the assessments were made on all plants. For the assessment of the uniformity of vegetatively propagated varieties, a population standard of 1% and an acceptance probability of at least 95% were adopted.

Table 1: Accessions used for the study

S. No.	Accessions	S. No.	Accessions	S. No.	Accessions
1	Acc. Js- 1	13	Acc. Js- 15	25	Acc. Js- 27
2	Acc. Js- 2	14	Acc. Js- 16	26	Acc. Js- 28
3	Acc. Js- 3	15	Acc. Js- 17	27	Acc. Js- 29
4	Acc. Js- 4	16	Acc. Js- 18	28	Acc. Js- 30
5	Acc. Js- 5	17	Acc. Js- 19	29	Acc. Js- 31
6	Acc. Js- 6	18	Acc. Js- 20	30	Acc. Js- 32
7	Acc. Js- 8	19	Acc. Js- 21	31	Acc. Js- 33
8	Acc. Js- 9	20	Acc. Js- 22	32	Acc. Js- 34
9	Acc. Js- 11	21	Acc. Js- 23	33	Acc. Js- 35
10	Acc. Js- 12	22	Acc. Js- 24	34	Acc. Js- 36
11	Acc. Js- 13	23	Acc. Js- 25		
12	Acc. Js- 14	24	Acc. Js- 26		

The Table 2 consists of the 40 characters, their codes and descriptor states which had been recorded. Scoring has been given from a range of 1 to 9 for the 40 qualitative characters of the 34 genotypes. A single measurement of a number of individual plants or parts of plants (MS) had been taken for the characters such as plant height (at flowering), number of forks per cyme, size of calyx lobes, Petiole length and flower bud length. A single observation of a group of plants or part of plants (VG) had been visually assessed for the characters like plant growth type, plant growth habit, young shoot anthocyanin colouration (shoots up to 30 cm from growing tip), young shoot intensity of anthocyanin colouration, ridges on the stem, leaf arrangement/ phyllotaxy, leaf size, intensity of green colour (upper side of mature leaf), leaf anthocyanin colouration, leaf glossiness on upper side (mature leaf), leaf blade undulations, leaf margin undulation, leaf type, shape of leaf blade – simple leaf, leaf tip, petiole color, flower bearing habit, flower bearing- position, boldness of flower bud, flower bud colour, tinge on flower bud, flower colour on opening, flower shape, shape of corolla lobe, flower petal tip, reflexing of petal whorls, reflexing of petal tips and margins and seed setting. Observations of individual plants or parts of plants (VS) had been visually assessed for the following characters like shape of base of leaf blade, flower type, flower bud shape, stipules, petiole, calyx and root suckers.

For the Principal Component Analysis (PCA), the PAST 3 application was used. Principal Components (PCs) with eigenvalue of more than one were selected as stated by Jeffers (1967) [7] and standardized values were used to conduct the PCA. Using the scree plot, the major contributors of variation were visually assessed. Agglomerative Hierarchical Clustering (AHC) and correlation matrix were constructed for all the 40 characters using the PAST 3 application.

Table 2: DUS descriptors used for the morphological characterization of the *Jasminum sambac* genotypes

S. No.	Characters	Code	Descriptor states
1	Plant growth type	PGT	Shrub/ Climber
2	Plant height (at flowering)	PH(F)	Short/ Medium/ Tall
3	Plant growth habit	PGH	Upright/ Semi upright/ Intermediate/ Spreading/Strongly spreading
4	Young shoot anthocyanin colouration (Shoots up to 30 cm from growing tip)	YSA	Absent/ Present
5	Intensity of young shoot anthocyanin colouration	IYSA	Absent/ Weak/ Medium/ Strong
6	Ridges on the stem	RIS	Absent/ Present
7	Leaf arrangement/ Phyllotaxy	LA	Opposite/ Alternate
8	Leaf Type	LT	Simple/ Compound/ Trifoliate/ Pinnate
9	Leaf size	LS	Small/ Medium/ Large
10	Intensity of green colour (upper side of mature leaf)	IGC	Light/ Medium/ Dark
11	Leaf anthocyanin colouration	LAC	Absent/ Present
12	Leaf glossiness on upper side (mature leaf)	LG	Absent/ Present
13	Leaf blade undulations	LBU	Absent/ Present
14	Leaf margin undulation	LMU	Absent/ Weak/ Medium/ Strong
15	Shape of leaf blade – Simple leaf	SLB	Lanceolate/ Elliptic/ Ovate/ Circular
16	Leaf tip	LTIP	Sharp/ Medium/ Blunt
17	Shape of base of leaf blade	SBLB	Acute/ Obtuse/ Rounded/ Cordate/ Asymmetric
18	Stipules	STP	Absent/ Present
19	Petiole	PTL	Absent/ Present
20	Petiole length	PTLL	Short/ Medium/ Long
21	Petiole color	PTLC	Yellow green/ Light green/ Dark green/ Purple
22	Flower bearing habit	FBH	Solitary/ Cluster/ Both
23	Flower bearing- Position	FBP	Terminal/ Axillary/ Both
24	Number of forks per cyme	NFPC	Less/ Intermediate/ More
25	Calyx	CLX	Conspicuous/ Inconspicuous
26	Calyx lobes	CLXL	Short/ Medium/ Long

27	Flower bud length	FBL	Short/ Medium/ Long
28	Boldness of flower bud	BFB	Thin/ Medium/ Bold
29	Flower bud shape	FBS	Round and Short/ Round and Long/ Round and flattened/ Pointed and Short/ Pointed and Long
30	Flower bud colour	FBC	Pure white/ Off white/ Pink tinged/ Green tinged/ Yellow
31	Tinge on flower bud	TFB	Absent/ Present
32	Flower colour on opening	FCO	Pure white/ Off white/ Pink/ Yellow
33	Flower shape	FS	Rounded/ Star shaped
34	Shape of corolla lobe	SCL	Rounded/ Lanceolate
35	Flower petal tip	FPT	Blunt/ Sharp
36	Reflexing of petal whorls	RPW	Absent/ Present
37	Reflexing of petal tips and margins	RPTM	Absent/ Present
38	Flower type	FT	Single/ Semi-double/ Double/ Multi whorled
39	Seed setting	SS	Absent/ Present
40	Root suckers	RS	Absent/ Present

Results and Discussion

To assess the variability of 34 *Jasminum sambac* genotypes collected from all over Tamil Nadu, principal component analysis was performed. Principal component analysis is a widely used dimensionality-reduction method for multivariate data while preserving as much data as possible. PCA is used to identify a minimum number of components which accounts for maximum variability out of the total variability (Anderson, 1972). PCA also helps in ranking the genotypes based on the PC scores.

Variability

PCA test in 34 genotypes for all the 40 characters resulted in fifteen Principal Components (PCs) with an eigenvalue more than one (Table 3). 97.21 per cent was the cumulative variability exhibited by these fifteen PCs. Out of these fifteen PCs, the first three PCs expressed more variability of 59.32 per cent in the proposed characters. PC1, PC2 and PC3 expressed 30.65 per cent, 15.78 per cent and 12.89 per cent variability respectively among the germplasm for the 40 traits under study (Table 3).

Table 3: Eigen value, percentage of variance and cumulative variance of *Jasminum sambac* genotypes

Principal component	Eigenvalue	% Variance	Cumulative variance
1	40.1095	30.651	30.651
2	20.653	15.783	46.434
3	16.8681	12.89	59.324
4	12.4563	9.5189	68.8429
5	8.12431	6.2084	75.0513
6	5.56309	4.2512	79.3025
7	4.74339	3.6248	82.9273
8	4.07348	3.1129	86.0402
9	3.4231	2.6159	88.6561
10	2.70067	2.0638	90.7199
11	2.36829	1.8098	92.5297
12	1.82857	1.3974	93.9271
13	1.514	1.157	95.0841
14	1.50153	1.1474	96.2315
15	1.27947	0.97775	97.20925

Scree Plot

Based on Eigenvalues, a Scree plot was constructed with the components on X-axis and eigenvalues on Y-axis which explains the percentage of variance associated with each PC (Fig.1). The PC1 shows a maximum variability of 30.65 per cent with an eigenvalue of 40.11. The variance then gradually decreased herewith Nachimuthu *et al.* (2014) [8]. A semi-curve line was observed after 13th PC. After 16th PC the curve gradually begins to flatten with little variation. From the **Fig. 1** it is clear that the selection PC1, PC2 and PC3 among the 33 PCs will result in maximum variability.

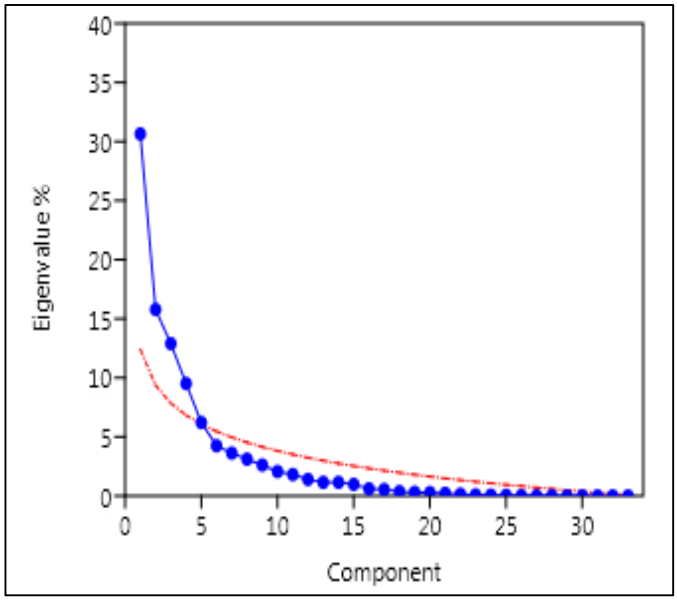
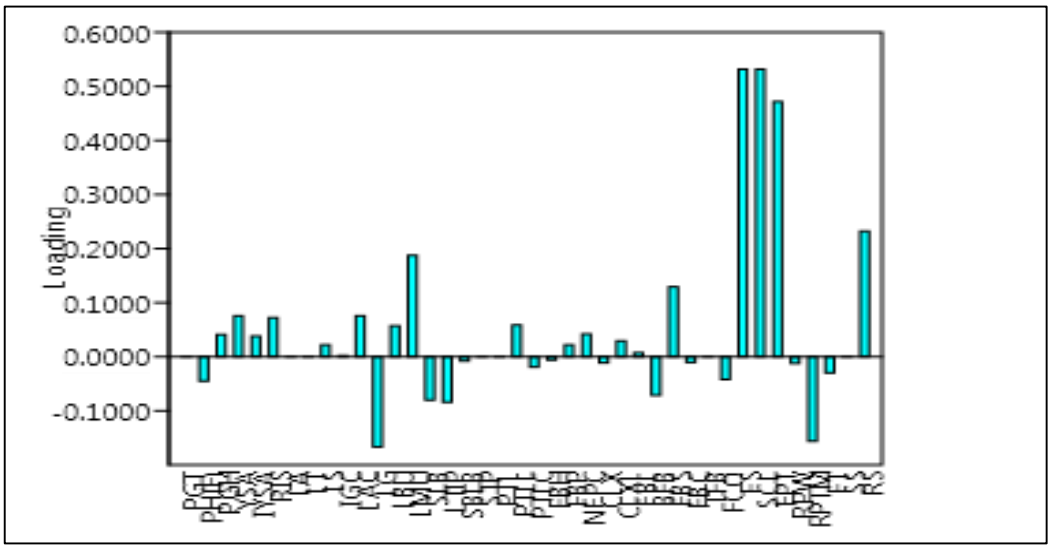


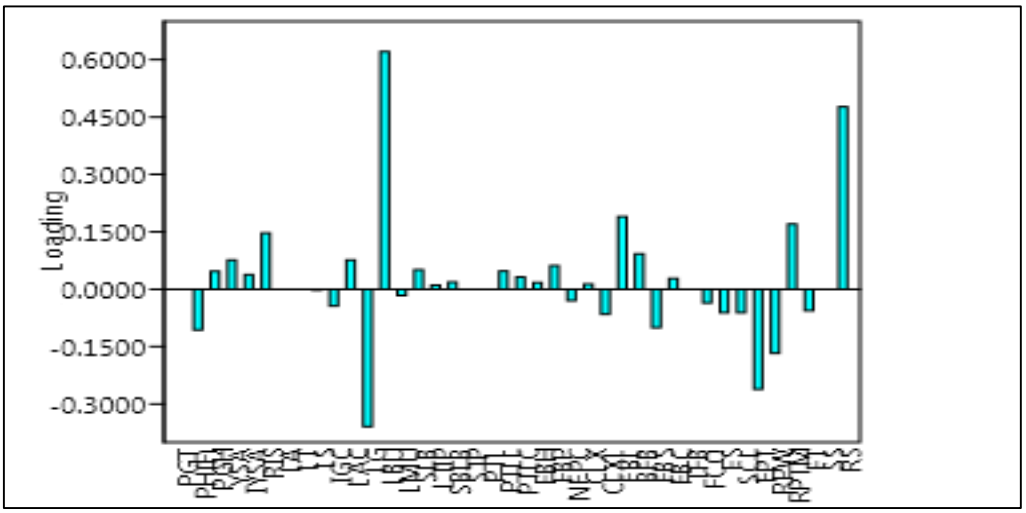
Fig 1: Scree plot of PCA of *Jasminum sambac* genotypes between eigenvalue and components

Loading plot:

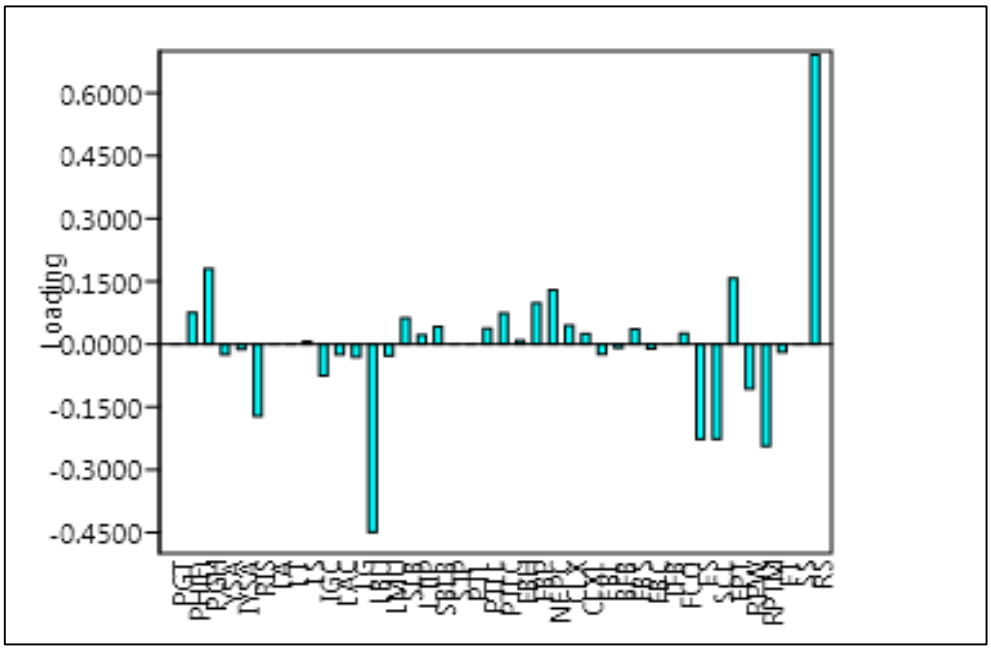
Graphical representations of Loading plot for PC1, PC2 and PC3 (Fig. (2a), (2b), (2c)) were extracted from the PCA. This led to the estimation of characters that contribute maximum variability out of the total variability. The characters with a loading score of > 0.5 were selected from each principal component. From the Loading plot PC1, it is clear that majority of the floral characters such as flower bud shape, flower shape, shape of corolla lobe, flower petal tip and one leaf character i.e., leaf margin undulation comes under PC1 (Table 4). From the Loading plot PC2, it can be seen that the characters such as leaf blade undulations, flower bud length and root suckers comes under PC2 and in PC3 only one root character i.e., root suckers come under PC3 (Table 4). These are the characters with maximum variation in each component.



(2a) PC 1



(2b) PC 2



(2c) PC 3

Fig. 2a, 2b, 2c: Graphical representation of loading plot for different characters in PC1, PC2 and PC3 respectively.

Table 4: Selection of traits having values > 0.5 in each PC for the interpretation of PCA.

	PC 1	PC2	PC3
Traits	Leaf margin undulation Flower bud shape Flower shape Shape of corolla lobe Flower petal tip	Leaf blade undulations Flower bud length Root suckers	Root suckers

Genotype selection on the basis of PC scores

Principal component analysis done on 34 genotypes of *Jasminum sambac* resulted in the estimation of genotypes with maximum variability out of the total variability in each principal component. Genotypes were selected based on the PC scores with positive values > 1.0 in each principal component (Table 5). In PC1, the positive values > 1.0 range from 1.5423 (Acc. Js- 15) to 12.546 (Acc. Js- 32). In PC2, the positive values > 1.0 range from 1.0587 (Acc. Js- 4) to 10.047 (Acc. Js- 25). In PC3, the positive values > 1.0 range from 1.5187 (Acc. Js- 20) to 7.9642 (Acc. Js- 12). It can be seen from the Table 5 that Acc. Js- 24, Acc. Js- 25 and Acc. Js- 32 comes under both PC1 and PC2, Acc. Js- 14, Acc. Js- 20, Acc. Js- 27 and Acc. Js- 30 comes under both PC2 and PC3 and Acc. Js- 11, Acc. Js- 12, Acc. Js- 13, Acc. Js-15 and Acc. Js- 23 comes under both PC1 and PC3. This shows that these above mentioned genotypes differ in the characters coming under two principal components (Table 4). From this we can conclude that the genotypes given in the Table 5for each PC shows maximum variability for the characters given in Table 4 for each PC.

Table 5: Selection of genotypes having values > 1.0 in each component on the basis of PC score

PC1	PC2	PC3
Acc. Js- 11 (11.757) Acc. Js- 12 (1.9027) Acc. Js- 13 (12.145) Acc. Js- 15 (1.5423) Acc. Js- 16 (8.8005) Acc. Js- 23 (11.254) Acc. Js- 24 (10.14) Acc. Js- 25 (8.7815) Acc. Js- 26 (7.9901) Acc. Js- 32 (12.546) Acc. Js- 34 (4.551)	Acc. Js- 4 (1.0587) Acc. Js- 6 (5.2865) Acc. Js- 14 (2.3012) Acc. Js- 19 (5.0356) Acc. Js- 20 (8.5131) Acc. Js- 21 (5.3624) Acc. Js- 22 (1.2329) Acc. Js- 24 (2.6281) Acc. Js- 25 (10.047) Acc. Js- 27 (4.0197) Acc. Js- 30 (1.9243) Acc. Js- 31 (6.5455) Acc. Js- 32 (6.5022) Acc. Js- 36 (1.6364)	Acc. Js- 11 (4.1102) Acc. Js- 12 (7.9642) Acc. Js- 13 (4.9026) Acc. Js- 14 (6.1683) Acc. Js- 15 (5.6651) Acc. Js- 17 (3.8412) Acc. Js- 20 (1.5187) Acc. Js- 23 (2.1018) Acc. Js- 27 (5.3085) Acc. Js- 30 (4.99) Acc. Js- 33 (4.5048) Acc. Js- 35 (4.2994)

Scatter plot:

Scatter plot graph constructed using component 1 and 2 indicates clustering among the 34 genotypes clearly (Fig. 3a). The genotypes Acc. Js- 4, Acc. Js- 8, Acc. Js- 9, Acc. Js- 11, Acc. Js-13, Acc. Js- 16, Acc. Js- 20, Acc. Js- 21, Acc. Js- 25, Acc. Js- 31 and Acc. Js- 32 occupied the convex hulls. From the result obtained from the scatter plot of PC 1 and PC2, the maximum variation was observed for the characters such as leaf blade undulation, root suckers, leaf glossiness on the upper side of the leaf, flower shape, shape of corolla lobe, flower petal tip and reflexing of petal tips and margins. The genotypes such as Acc. Js- 11, Acc. Js- 12, Acc. Js- 13, Acc. Js- 14, Acc. Js- 16, Acc. Js- 20, Acc. Js- 25, Acc. Js- 26, Acc. Js- 27 and Acc. Js- 34 occupied the convex hulls of the scatter plot constructed between PC2 and PC3. From the result obtained from the scatter plot of PC2 and PC3, the maximum variation was observed for the characters such as root suckers, leaf blade undulation, leaf glossiness, flower petal tip, reflexing of petal tips and margins, reflexing of petal whorls, flower bud length and plant growth habit. The characters other than this did not show much diversity

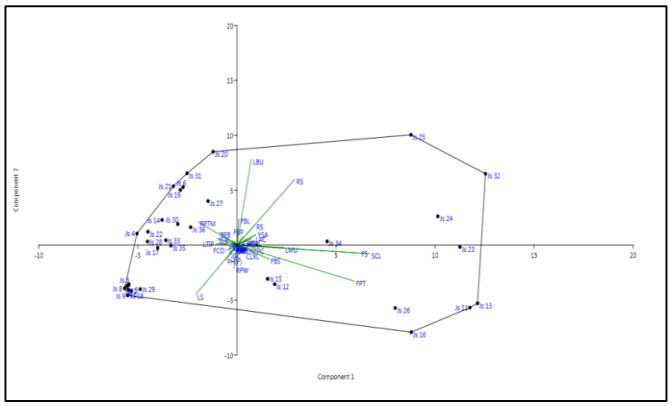


Fig 3a: Scatter plot between PC1 and PC2

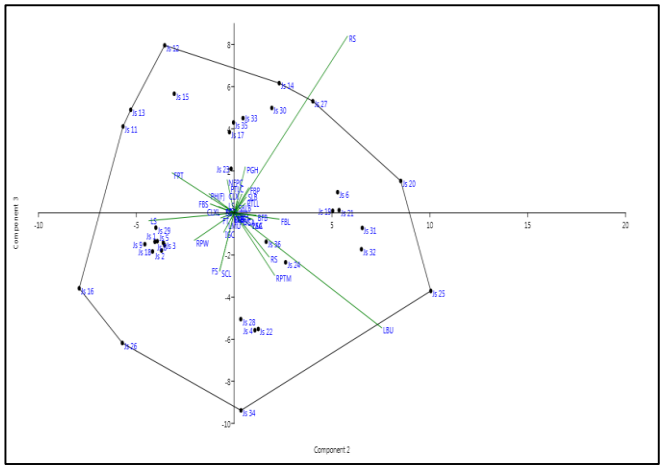


Fig 3b: Scatter plot between PC2 and PC3

Correlation

Assessment of correlation among the characters aid in designing hybridization programmes for the crop development (Popoola *et al*., 2016). This is because correlation of morphological traits between the parental lines will be reflected in the hybrid lines too. From the Fig. 4, it can be seen that there were many positively correlated traits out of which leaf anthocyanin coloration, young shoot anthocyanin coloration, tinge on flower buds and intensity of young shoot anthocyanin coloration are highly positively correlated. Another trait, flower shape and shape of corolla lobe are also positively correlated which makes sense.

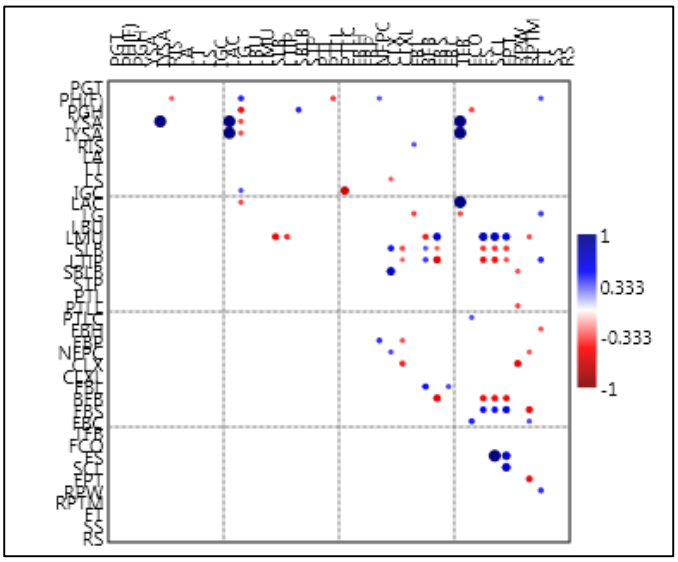


Fig 4: Correlation plot between different traits

Clustering

The dendrogram obtained through Agglomerative Hierarchical Clustering (AHC) analysis using Unweighted Pair Group Method with Arithmetic mean (UPGMA) resulted in two major clusters. The Cluster A consists of two genotypes. The Cluster B consists of all the other 32 genotypes. Acc. Js- 25 and Acc. Js- 32 having similar leaf margin undulation, flower bud shape, flower shape, shape of corolla lobe, flower petal tip, leaf blade undulations, flower bud length and root suckers were in same cluster A. In the Cluster B, the genotypes Acc. Js- 11, Acc. Js- 13, Acc. Js- 16, Acc. Js- 23, Acc. Js- 24, Acc. Js- 26 and Acc. Js- 34 having similar flower bud shape, flower shape, shape of corolla lobe, flower petal tip comes under the same sub- cluster. The genotypes Acc. Js- 12 and Acc. Js- 15 having root suckers were placed in a sub- cluster of Cluster B. The genotypes Acc. Js- 14, Acc. Js- 20, Acc. Js- 27, Acc. Js- 30 and Acc. Js- 36 having similar flower bud length were placed under a sub-cluster of Cluster B. Hence genotypes having similar characters were grouped under each clusters and the Cluster A and B shows maximum variability among the 40 qualitative traits. Thus the genotypes Acc. Js- 11, Acc. Js- 12, Acc. Js- 13, Acc. Js- 14, Acc. Js- 20, Acc. Js- 25, Acc. Js- 27 and Acc. Js- 32 were found to be the most diverse genotypes.

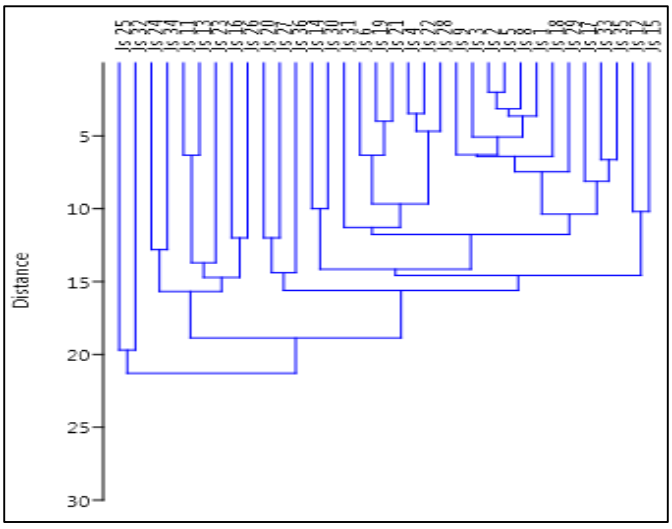


Fig 5: Agglomerative Hierarchical Clustering

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