

Morphological characterization and principal component analysis of cowa (*Garcinia cowa* Roxb.) germplasm

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ABSTRACT

Garcinia cowa Roxb. is a neglected underutilized minor fruit in Bangladesh. There are several types of Cowa found in different locations of the country. Therefore, it is necessary to investigate the variations among different types of Cowa. To do so, five Cowa accessions were collected from Barishal and Patuakhali districts of Bangladesh to analyze the morphological variations among the germplasm and also conducted the principal component analysis (PCA) to determine the ideal amount of morphological characteristics that account for a large portion of variability. Morphological traits viz., leaf shape, base, apex, length, width, petiole length, diameter, flower characters, weight of fruit, length of fruit, diameter of fruit, seed number per fruit, length of seed, diameter and weight of seed were recorded. Wide variations were observed in leaf, fruits and seed characters of different Cowa germplasm. The longest leaves (15.70 cm), maximum fruit weight (30.56 g), highest fruit length (4.43 cm), diameter (3.90 cm) and highest seed weight (7.42 g) were found in ACS GC4. The number of seeds per fruit was minimum in case of ACS GC3. PCA for 17 morphological characters of Cowa elucidated that four principle components with eigen values more than one accounted for 100 per cent of the total variability and the maximum variation was exhibited by the leaf length which accounted for 91.146%. From genotypic scatter plot, it was noticed that ACS GC3 has the highest variability as compared to other four germplasm. Based on the PCA and genotypic scatter plot, germplasm ACS GC3 can be a potential material for future varietal improvement program.

Keywords: Cowa germplasm, morphological traits, principal component analysis

INTRODUCTION

Cowa (*Garcinia cowa* Roxb.) is a tropical and subtropical evergreen wild edible fruit that is a member of the Clusiaceae family (or Guttiferae). More than 250 species of evergreen, lactiferous, dioecious trees and shrubs are included in the genus *Garcinia*, which is found in moist, lowland tropical forests (Sweeney, 2008). The tree of Cowa can be 5-10 meters tall with several branches and green leaves. In the context of Bangladesh, this fruit is frequently referred to as Kawphal or Kao-gola, among other local names. The fruit in question, sometimes referred to as a “cowboy fruit,” is considered to be of low significance or underutilized within the agricultural context of Bangladesh (Ashrafuzzaman *et al.*, 2021).

The utilization of various components of Cowa, including its roots, leaves, and dried fruits, as traditional remedies is prevalent in several regions of India. The Cowa fruit possesses a distinct acidic taste and exhibits astringent properties that have been traditionally employed in the management of dysentery. Cowa fruit is recognized as a significant and valuable reservoir of bioactive chemicals, which have been traditionally employed for the therapeutic management of wounds and ulcers. It has been found that cowa fruit possesses various additional bioactive components with anti-inflammatory and antioxidant properties (Panthong *et al.*, 2009).

According to the report of Tripathi (2021), the fruit of *Garcinia cowa* possesses significant medicinal properties. The researchers successfully

identified and described a collection of compounds, including an uncommon class of polyprenylated acylphloroglucinol derivatives, as well as three further novel garcicowins and nine previously identified analogues. These compounds were extracted from the twigs of *Garcinia cowa*. They tested the cytotoxicity of the derived compound and their toxicity toward the cancer cells were demonstrated selectively. Cow fruit is a vital source of minerals particularly Na and Fe and it contains 29.41mg/100g DW Na and 4.56 mg/100g DW Fe (Hossain *et al.*, 2021).

The plant species Cowa is found in its native habitat along the roadside and inside the forests of various districts in Bangladesh, including Chottogram Hill Tracts, Cox's Bazar, Barisal, Sylhet, Patuakhali, Bagerhat and Moulavibazar. Cowa germplasm's morphological traits somewhat different to some extent. Therefore, this study was conducted to collect different Cowa germplasm from two locations, conserved them at the BAU-GPC (Bangladesh Agricultural University Germplasm Center) and investigate the morphological traits of leaves, flowers, fruits and seeds. Principal Component Analysis of all studied traits were accomplished to find out a germplasm with higher variability.

MATERIALS AND METHODS

The morphological characters of *Garcinia cowa* Roxb. were recorded from mature and bearing trees. A total of five accessions of Cowa were collected from three locations of Bangladesh namely BAU-GPC, Mymensingh, Barishal and Patuakhali district. Leaves, flowers, fruits were collected from the experimental site to the Department of Horticulture, Bangladesh Agricultural University for investigation. Length, diameter of leaves, fruits, seeds were measured by using measuring tape and slide calipers, respectively. Weight of the sample was recorded by digital balance. Data were statistically analyzed using Statistix 10.0. The Duncan Multiple Range Test (DMRT) was employed to determine the statistically significant differences between two mean values at significance levels of 5% and 1%. The use of Principal Component Analysis (PCA) was carried out using Ward's approach.

RESULTS AND DISCUSSION

Morphological traits of leaves

Leaf traits of different germplasm slightly varied among each other. Leaf shape of two germplasm were elliptically lanceolate, rest two germplasm were lanceolate and one germplasm was acuminate. Leaf apex was obtuse in ACS GC1 while all other germplasm showed acuminate leaf apex. The base of leaves was cuneate, rounded and acute (Table 1, Plate 1 and 2).

Significant variations were observed in the measurements of leaf length, leaf breadth, petiole length, petiole diameter, and leaf area among different germplasm samples of Cowa. The leaf length measurements revealed that ACS GC4 exhibited the greatest value of 15.70 cm, followed by ACS GC2 with a length of 14.80 cm, ACS GC3 with a length of 14.43 cm, and ACS GC5 with the lowest length of 12.53 cm, as presented in Table 2. The maximum leaf width observed in ACS GC1 was 4.83 cm and the minimum width recorded in ACS GC3 (4.10cm). The petiole length of leaves varied from 0.87 cm to 1.07 cm while petiole diameter varied from 0.18 cm to 0.20 cm (Table 2). In case of leaf area, a wide variation was observed among the Cowa germplasm and the range was 36.74cm² to 56.66cm² (Table 2).

Morphological traits of flowers

The flowers exhibited dioecy, with a tiny size and a yellow coloration. Notably, the male flowers were comparatively smaller in size when compared to the female flowers. Additionally, the flowers were found to be either axillary or terminal in their arrangement. The male flowers exhibited yellow petals, with a base characterized by four bracts. Additionally, the flowers possessed 40-50 anther filaments that formed a cohesive mass. Female flowers were light-yellow color, usually solitary, axillary, larger than male, flower buds were round in shape. The length of buds exhibited variation between the different accessions of Cowa. The maximum bud length was seen in ACS GC1 and ACS GC4, measuring 1.23 cm, while the minimum length was recorded in ACS GC3 and ACS GC5, measuring 1.03 cm (Table 3). The highest sepal length was observed in ACS GC4 (0.63 cm), while the lowest sepal length was observed in ACS GC5

Table 1: The shape, apex, and base of cowa leaf according to the germplasm

Germplasm	Leaf shape	Leaf apex	Leaf base
ACS GC1	Elliptically lanceolate	Obtuse	Cuneate
ACS GC2	Elliptically lanceolate	Acuminate	Rounded
ACS GC3	Lanceolate	Acuminate	Cuneate
ACS GC4	Lanceolate	Acuminate	Cuneate
ACS GC5	Acuminate	Acuminate	Acute

Table 2: The measurements of length and breadth of leaf as well as petiole and leaf area for the Cowa germplasm.

Germplasm	Leaf length (cm)	Leaf diameter (cm)	Petiole length (cm)	Petiole diameter (cm)	Leaf area(cm ²)
ACS GC1	13.87 c	4.83 a	0.87 c	0.19 b	50.07c
ACS GC2	14.80 b	4.70 a	1.03 ab	0.19 b	52.51b
ACS GC3	14.43 b	4.10 d	0.93 bc	0.18 c	47.16d
ACS GC4	15.70 a	4.53 b	1.07 a	0.20 a	56.66a
ACS GC5	12.53 d	4.27 c	0.93 bc	0.18 c	36.74e
LSD (0.05)	0.46	0.16	0.11	0.09	0.99
Level of Sig.	**	**	**	**	**
CV(%)	1.71	1.84	5.31	2.77	6.12

** indicates significant at 1% levels of probability

Table 3: The flower features of Cowa germplasm.

Germplasm	Bud length (cm)	Sepal length (cm)	Sepal width (cm)	Petal length (cm)	Petal width (cm)
ACS GC1	1.23 a	0.54 b	0.24 b	1.17 a	0.60 b
ACS GC2	1.07 b	0.60 a	0.24 b	1.03 b	0.70 a
ACS GC3	1.03 b	0.42 c	0.30 a	1.23 a	0.62 b
ACS GC4	1.23 a	0.63 a	0.31 a	1.27 a	0.73 a
ACS GC5	1.03 b	0.42 c	0.22 b	0.87 c	0.52 c
LSD (0.05)	0.11	0.05	0.03	0.11	0.05
Level of Sig.	*	**	**	**	**
CV(%)	5.57	5.31	6.14	3.78	6.14

*&** indicates significant at 5% and 1% levels of probability

Table 4: Fruit and seed traits of Cowa germplasm

Germplasm	Fruit fresh weight (g)	Fruit length (cm)	Fruit width (cm)	No. of seeds/ fruit	Seed weight (gm)	Seed length (cm)	Seed diameter (cm)
ACS GC1	24.09c	3.54c	3.75b	6.00b	6.12c	2.16c	1.14bc
ACS GC2	24.82c	3.52c	3.68b	7.33a	7.11b	2.06c	1.08c
ACS GC3	28.66b	4.18b	3.81ab	4.00d	6.96b	2.55a	1.40a
ACS GC4	31.29a	4.43a	3.90a	7.33a	7.42a	2.38b	1.20b
ACS GC5	18.26d	3.11d	3.37c	5.00c	5.21d	1.76d	0.79d
LSD 0.05	1.16	0.18	0.15	0.73	0.26	0.15	0.08
Level of sig	**	**	**	**	**	**	**
CV(%)	6.53	2.51	2.42	3.72	3.92	2.07	3.92

** Significant at 1% level of significance

Table 5: Eigen value and percentage of variance for corresponding 17 component characters of 5 Cowa germplasm

Components	Initial Eigenvalues Total	% of Variance	Cumulative %
Leaf length (cm)	78.216	91.146	91.146
Leaf width (cm)	7.056	8.222	99.369
Leaf petiole length (cm)	0.421	0.491	99.860
Leaf petiole width (cm)	0.120	0.140	100.000
Leaf area (cm ²)	0.000	0.000	100.000
Bud length (cm)	0.000	0.000	100.000
Length of sepal (cm)	0.000	0.000	100.000
Width of sepal (cm)	0.000	0.000	100.000
Length of petals (cm)	0.000	0.000	100.000
Width of petals (cm)	0.000	0.000	100.000
Fruit weight (gm)	0.000	0.000	100.000
Fruit length (cm)	0.000	0.000	100.000
Fruit width (cm)	0.000	0.000	100.000
No. of seed/fruit	0.000	0.000	100.000
Seed weight (gm)	0.000	0.000	100.000
Seed length (cm)	0.000	0.000	100.000
Seed width (cm)	0.000	0.000	100.000

Table 6: Score of Cowa germplasm

Germplasm	PC 1	PC 2	PC 3	PC 4
ACS GC1	-0.28474	0.82323	2.1076	-0.16935
ACS GC2	0.59293	1.7345	-1.0169	-0.99438
ACS GC3	0.59333	-3.4471	-0.12362	-0.3319
ACS GC4	4.1596	0.62389	-0.3924	0.901
ACS GC5	-5.0612	0.26553	-0.57471	0.59463

(0.42 cm) (Table 3). The cow accessions exhibited variation in sepal breadth as well. Table 3 displays the observation that ACS GC4 had the highest sepal width measurement at 0.31 cm, while ACS GC5 exhibited the lowest measurement at 0.22 cm.

The petal length of Cowa accessions was observed to exhibit variation among the accessions. The findings of the study indicate that the petal length of ACS GC4 was the largest, measuring 1.27 cm. This measurement was statistically comparable to the petal lengths of ACS GC3 (1.23 cm) and ACS GC1 (1.17 cm). Conversely, the lowest petal length was observed in ACS GC5, measuring 0.87 cm (Table 3). Variation in petal breadth was seen among the different accessions. The observation revealed that ACS GC4 exhibited the greatest petal width, measuring 0.73 cm, which was comparable to ACS GC2 with a petal width of 0.70 cm.

Following these, ACS GC3 displayed a petal width of 0.62 cm, ACS GC1 had a petal width of 0.60 cm, and ACS GC5 exhibited the lowest petal width at 0.52 cm (Table 3).

Morphological traits of fruits

Variation in fruit shape was observed among the Cowa germplasm. Among the five germplasm, fruit shape of three germplasm (ACS GC1, ACS GC2 and ACS GC5) was rounded and the rest two germplasm (ACS GC 3 and ACS GC 4) found globose shape. Fruit surface color also varied from brownish to yellow (Plate 3). Fruit flesh color varies from light orange to orange (Plate 4).

The fresh weight of the fruit was assessed subsequent to harvesting, revealing variations in fruit weight among different accessions. The fruit weight analysis revealed that ACS GC4 had the

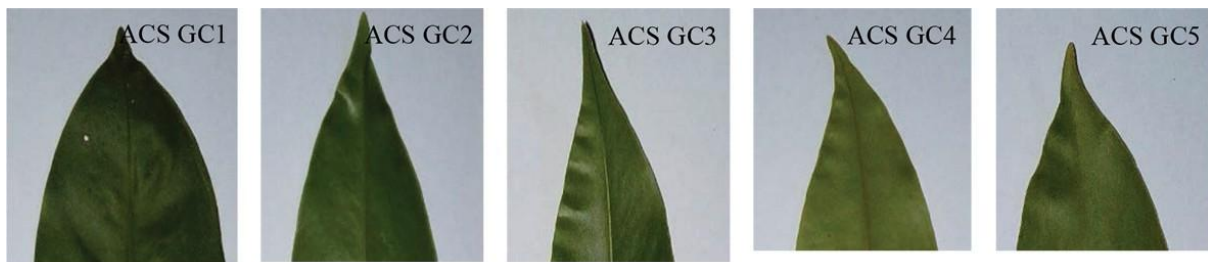


Plate 1. Leaf apex status of the Cowa germplasm



Plate 2. Leaf base status of the Cowa germplasm

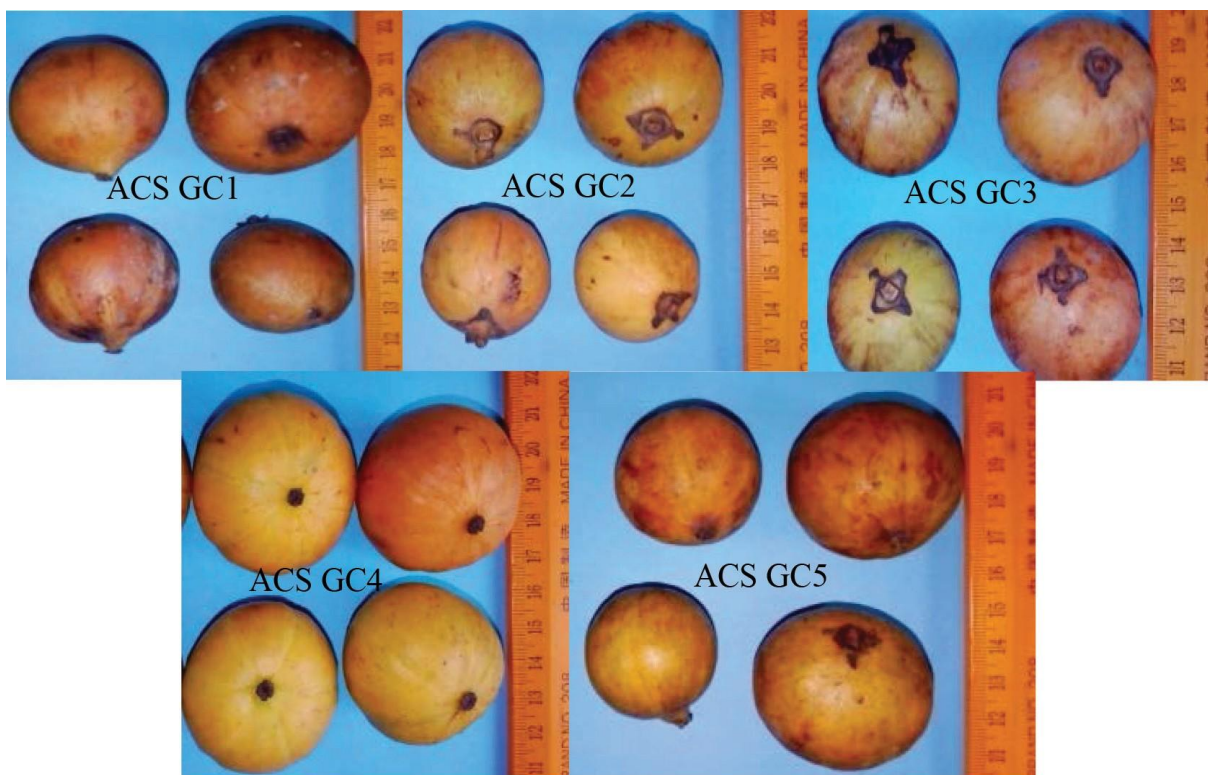


Plate 3. Fruit shape and surface color of different Cowa germplasm

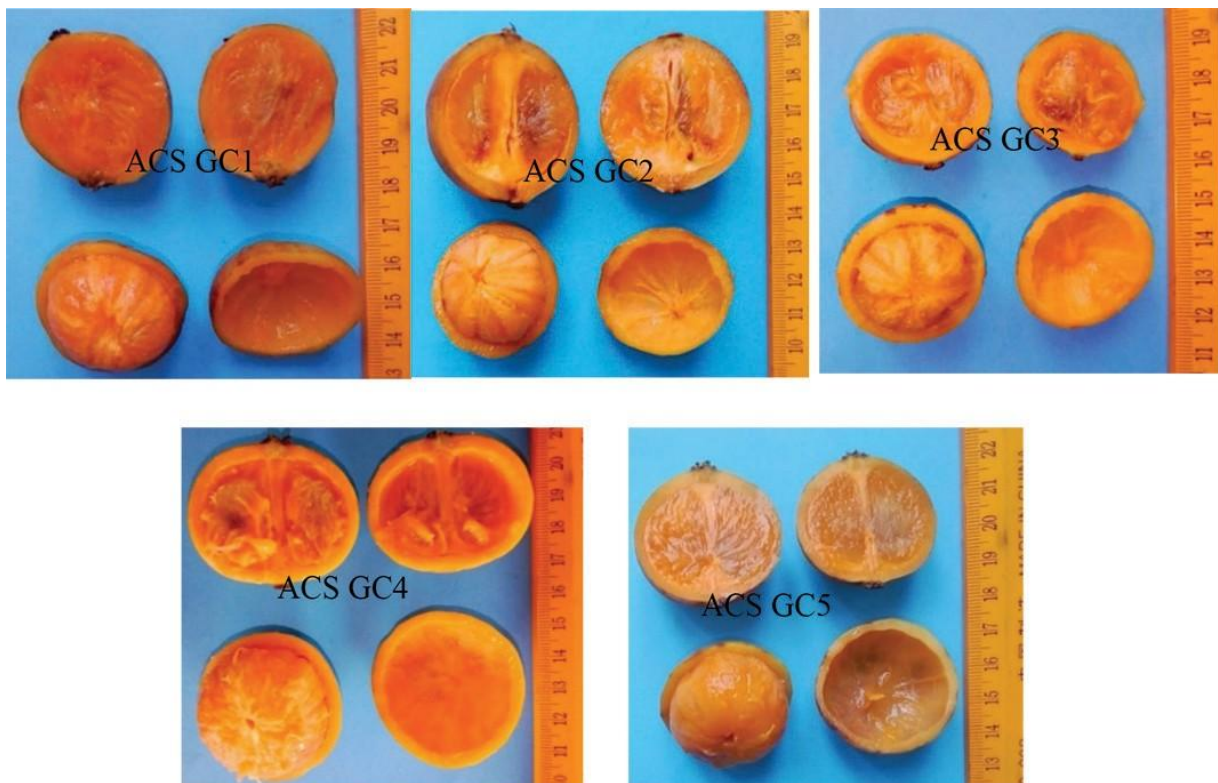


Plate 4. Flesh color of different Cowa germplasm

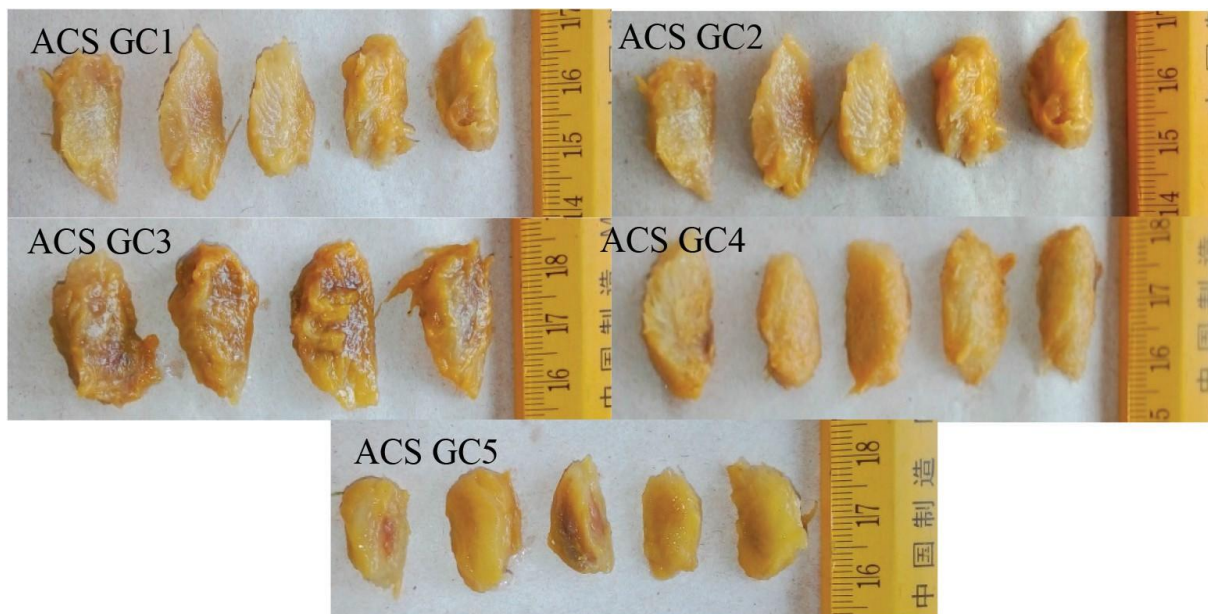


Plate 5. Seeds of different Cowa germplasm

Principal component Analysis of Cowa germplasm

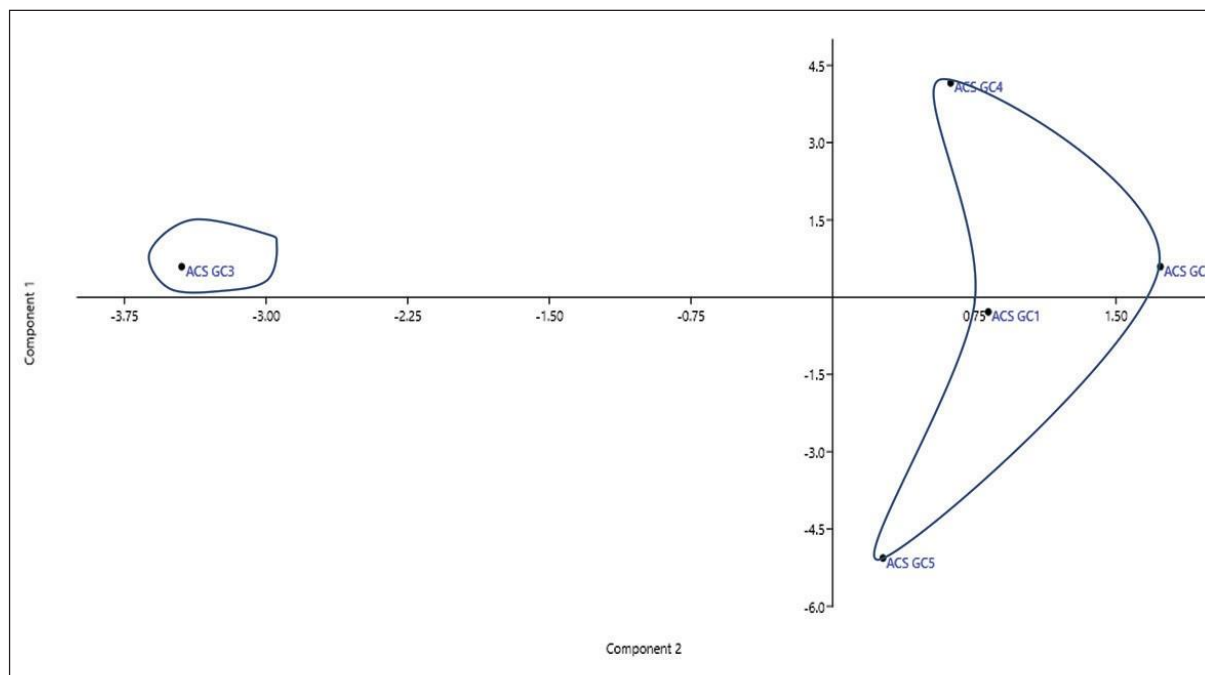


Fig. 1: Genotypic scatter plot illustrating the relationship between PC1 and PC2 germplasm

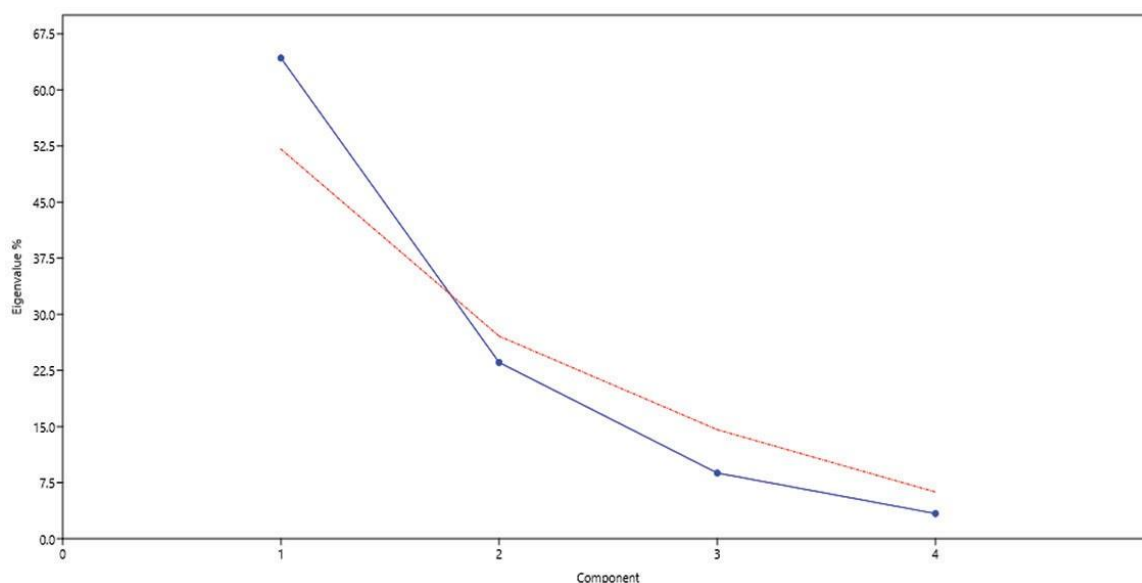


Fig. 2: Scree plot of Cowa germplasm

highest weight at 31.29 gm, followed by ACS GC3 at 28.66 gm, ACS GC2 at 24.82 gm, and ACS GC5 had the lowest weight at 18.26 gm (Table 4). There was considerable variation in fruit length among the accessions. The fruit length measurements revealed that ACS GC4 had the highest value at 4.43 cm, followed by ACS GC3 at 4.18 cm, ACS GC1 at 3.54 cm, and ACS GC5 had the lowest value

at 3.11 cm (Table 4). The fruit width of various Cowa accessions exhibited significant variation among each other. The observation revealed that ACS GC4 exhibited the greatest fruit width, measuring 3.90 cm. This was followed by ACS GC3 with a fruit width of 3.81 cm, ACS GC1 with a fruit width of 3.75 cm, and ACS GC5 with the smallest fruit width of 3.37 cm, as indicated in Table 4.

Morphological traits of seeds

The hue of Cowa seeds was predominantly yellowish, as depicted in Plate 5. The seed count per fruit exhibited variability throughout the germplasm. The highest number of seeds was observed in ACS GC2 and ACS GC4, with a recorded value of 7.33. This was followed by ACS GC1 with 6 seeds, ACS GC5 with 5 seeds, and the lowest number of seeds was found in ACS GC3 with 4 seeds (Table 4). The seed weight, length, and diameter exhibited variation within the germplasm. The recorded seed weights for ACS GC4, ACS GC2, ACS GC3, and ACS GC5 were 7.42 gm, 7.11 gm, 6.96 gm, and 5.21 gm, respectively, as shown in Table 4. In a similar vein, the dimensions of the seeds were observed to be greater in ACS GC3, with a length of 2.55 cm and a diameter of 1.40 cm. This was followed by ACS GC4, which exhibited a length of 2.38 cm and a diameter of 1.20 cm. ACS GC1 displayed slightly smaller seed dimensions, with a length of 2.16 cm and a diameter of 1.14 cm. Conversely, the smallest seed dimensions were observed in ACS GC5, with a length of 1.76 cm and a diameter of 0.79 cm. These findings are presented in Table 4 and visually depicted in Plate 5.

Four lower and higher inter-accessions distances between pairs of 5 different Cowa germplasm accessions. The greatest inter genotypic distance observed from principal coordinate analysis was 78.22, which was observed between accession ACS GC3 and ACS GC1, followed by distances of 7.06, 0.42 and 0.12 among the accessions ACS GC2, ACS GC4 and ACS GC5. The shortest observed distance of 0.12 was found between ACS GC2 and ACS GC4 germplasms, while a distance of 0.42 was observed between ACS GC2 and ACS GC1 germplasm (Fig.1).

The findings of the principal component analysis unveiled that the first principal axis; leaf length largely accounted for the variation among the accessions, which alone contributed 78.216% of the total variations (Table 5). The cumulative contribution of the initial four characters along the major component axis, which possess eigen values exceeding unity, encompassed the entirety of the overall variance among 17 characters describing 5 Cowa germplasm. The rest eight characters contributed remaining 0.0% of total variation.

Principal Component Analysis (PCA) can be employed to examine the morphological features of Cowa germplasm in order to infer insights into the underlying structure of variables or components as follows:

- **Component 1** correlates well with the variables leaf petiole length, leaf area, bud length, length of sepal, width of sepal, length of petals, width of petals, fruit weight, fruit length, fruit width, seed weight and seed length showing as major factor to affect overall yield of the selected germplasm.

- **Components 2, 3, 4 and 5** were strongly correlated with variables No. of seeds/fruit, leaf width (cm), leaf length (cm) and seed weight (gm) and as express them as stand-alone factor to affect yield as a yield contributing factors.

-Variables close to an axis correlate with that principal component; one may consider that axis is a combination of its neighboring variables.

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