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Assessing Genetic Variability of Snake Gourd (*Trichasanthes Cucumerina* Var. Anguina L.) Germplasm Through Morphological Characterization and Multivariate Analysis

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ABSTRACT

Genetic divergence analysis is important for selecting genetically varied parents from existing germplasm for a successful breeding program. An investigation with thirty-three snake gourd germplasm was evaluated to assess their diversity using several qualitative and quantitative characters. Of the qualitative characters maximum variations were observed in fruit shape, fruit curvature, fruit skin color, and fruit skin lustre. The highest quantitative variation was observed in yield per plot (CV-49.44%), which was influenced by the higher number of fruit yields per plant (CV-49.22%). The first principal component (PC1) explained 30.6% while the second PC showed 18.3% of the total variability among the evaluated snake gourd germplasm. The clustering study classified them into five groupings based on twelve quantitative charac-ters where the most accessions (11) were under cluster V followed by cluster II (10). The max-imum cluster mean value was observed in cluster IV for the characters of fruit length (36.44 cm), number of fruits per plant (11.60), fruit yield per plant (2.34 kg), and yield per plot (9.37 kg). The snake gourd germplasm such as N-170, LAH-68, AHI-99, R-1, KI-36, AC-423 and AC-187 performed as wide variations according to biplot analysis while AC-118, and AR-39, AHI-104, AHI-99, AC-187, AC-357 superior through cluster analysis. Finally, the identified desirable traits from the prominent germplasm can be used for advancing the high-yielding va-rieties.

INTRODUCTION

Snake gourd (*Trichasanthes cucumerina* var. anguina L.) is a cultivated monoecious annual vegetable crop. It is classified as the Cucurbitaceous family, with chromosome number, 2n=22 (Rashid *et al*., 2014). It originated in Southeast Asia and Australia and is now grown in tropical and subtropical cli-mates (Ahmed *et al*., 2022). It is considered a quick-growing summer vegetable in Bangladesh. Moreo-ver, it has tremendous export potential due to its excellent keeping quality (Podder *et al*., 2010). It has a significant amount of protein (0.5%). Minerals (0.5%), fiber (0.5%), and carbs (3.3) (Gopalan, 1982). The snake gourd yield is somewhat lower in Bangladesh than in other snake gourd producing countries such as India and Thailand. (Ara *et al*., 2015). The total area of snake gourd cultivation in Bangladesh was 397 acres while producing 510 tons (BBS, 2022) due to limitations of yielding varieties. A significant number of local cultivars are grown, but no one has recognition for using the varietal improvement pur-poses. Even, no substantial initiatives have been paid to raise the improvement of snake gourd. Besides, the lack of genetic diversity, low harvest index, poor crop management, increased weed competition, and susceptibility to biotic and abiotic stress affect snake gourd production (Srinives, 2006). However, it is urgent to improve the snake gourd variety in terms of yield, broader adaption, quality, insect-pest re-sistance, disease resistance, and genetic diversity. (Ferdous *et al*., 2011).

The characterization and estimation of the genetic diversity among locally collected germplasm can be used for the germplasm exploitation of breeding programs (Jahan *et al*., 2022). The multivariate statisti-cal methods are capable of analyzing multiple measurements of the genetic diversity of individual germplasm. In addition, morphological, biochemical, or molecular marker-based characterizations are used to identify the desirable traits of a germplasm (Suman *et al*., 2019). According to Sabaghnia, *et al*. (2014), multivariate procedures are needed to characterize, evaluate, and classification of germplasm from a large number of accessions. Moreover, Cluster analysis and PCA (principal component analysis) were considered to assess the genetic diversity among the genotype collections of various crop species (Jatav *et al*., 2022). This analysis can identify and categorize objects, individuals, or variables based on the similarity of their attributes (Jain & Patil, 2016). In many countries, research has been done on snake gourds. For example, Podder *et al*. (2016) have discovered important characters by heterosis and combin-ing ability analysis. Ara *et al*. (2015) studied only morphological characterization without diversity anal-ysis. Islam *et al*. (2022) identified potential parents for the hybridization of snake gourds. Similarly, Khan *et al*. (2016) studied genetic variability, heritability, and path coefficient analysis of snake gourd.

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However, recently numerous genotypes of snake gourd have been collected and preserved by PGRC, Bangladesh Agricultural Research Institute (BARI) from domestic and abroad. The huge number of germplasm steal remains out of characterization. Therefore, the present study was carried out to evaluate the characterization and genetic diversity of snake gourds for crop improvement programs through mul-tivariate analysis.

MATERIALS AND METHODS

Experimental Site

The experiment was carried out at the experimental field of the Plant Genetic Resources Center of the Bangladesh Agricultural Research Institute, Gazipur, during the Kharif season 2020. The site was lo-cated at 23°980 N latitude and 90°412 E longitude at an elevation of 16 meters above sea level. The ex-perimental field pH was 6 with silty clay soil.

Experimental Materials Methods and Other Operations

Thirty-three landraces were used in this experiment those were collected from different parts of Bangla-desh (Table 1). Landraces were collected from saline-prone areas (Noakhali, Satkhira, Patuakhali, and Bhola) drought-prone areas (Naogoa, Nilphamari), Hilly areas (Khagarachari, Chattagram), and other districts of Bangladesh. Before sowing seeds were soaked in water for 12 hours and treated with provex @ 2.5g kg^{-1}. The seeds were shown in the pot for germination on 4 April 2020. After germination twen-ty aged seedling was transplanted in the main plot on 24 April 2020. The unit plot size was 2.5 m x 2 m. Every unit plot was two pits. Two plants were planted in each pit. One germplasm represented one treatment. Recommended fertilizers dose was applied as 10 ton ha^{-1} cow-dung, Urea, TSP, MoP, Gyp-sum, $ZnSO_4$, and Borax 175-175-150-100-12.5 and 10 kg ha^{-1}, respectively (Krishi Projukti Hatboi, 2020). The full doses of decomposed cow dung, TSP, Gypsum, $ZnSO_4$, and Borax as well as one-third of urea and MoP were applied during the final land preparation. The remaining urea and MoP were treated as top dressing in four stages 15, 35, 55, and 75 days, and 10 days after tran The remaining urea and MoP were treated as a top dressing in four installments at 15, 35, 55, 75, and 10, 15 days after transplantation respectively. The trail was made with bamboo and plastic wire for the proper growth and development of the snake gourd plants. Watering and intercultural operations were done as necessary. A sex pheromone trap was hung in the field to control cucurbit fruit fly. Both qualitative and quant

Table 1: List of collected snake gourd landraces from different parts of Bangladesh

Sl No.	Landrace code	Collected location	Geographical location (decimal)
1.	N-102	Jashore	23.066416, 89.057444
2.	N-170	Tangail	24.600019, 90.033215
3.	AC-21	Dhaka	24.216980, 90.199678
4.	AC-46	Dhaka	23.783274, 90.266677
5.	AC-118	Narayanganj	24.799980, 90.216645
6.	AC-187	Narsingdhi	24.010111, 90.663324
7.	AC-357	Gazipur	23.966686, 90.050011
8.	AC-423	Dhaka	23.650020, 90.349871
9.	AR-39	Noakhali	22.853567, 91.129798
10.	AR-48	Satkhira	23.955227, 90.573778
11.	AR-188	Jashore	23.119313, 89.193129
12.	AR-261	Chattagram	22.365567, 91.891113
13.	AR-262	Chattagram	22.302054, 91.810089
14.	AR-276	Chattagram	22.417627, 91.901413
15.	IAH-68	Gazipur	24.066647, 90.533333
16.	IAH-294	Khagrachari	23.111689, 92.001891
17.	RAI-125	Bogura	24.858485, 89.404221
18.	AHI-99	Jashore	23.157419, 89.184060
19.	AHI-104	Jashore	23.167993, 89.190411
20.	AHI-118	Jashore	23.142424, 89.181142
21.	MAH-03	Narayanganj	23.678982, 90.479602
22.	MAH-45	Narayanganj	23.671750, 90.486125
23.	R-1	Jenaida	23.549580, 89.155518
24.	R-69	Jenaida	23.551300, 89.156853

25.	I-4	Khagarachari	23.029886, 91.846659
26.	SA-32	Nilphamari	25.939917, 88.825835
27.	KASI-34	Gopalganj	23.003076, 89.840033
28.	KI-36	Ishurdi	24.039581, 89.135425
29.	ATR-37	Gazipur	24.027079, 90.393373
30.	RIM-50	Ishurdi	24.051885, 89.134405
31.	MK-90	Naogoa	24.288048, 89.827102
32.	RISA-126	Bhola	22.685099, 90.640633
33.	TRMR-130	Comilla	23.577497, 91.138184

Statistical Analysis

The range, mean, standard deviation, and coefficient of variation were calculated in Office Ex-cel 2016. Moreover, the principal component analysis and the cluster analysis were analyzed by using the software R 4.3.1 (2023).

Passport Information and Data Recorded

Collector's number: The original number assigned by the collector of the sample is normally composed of the name or initials of the collector(s) followed by a number. This item is essen-tial for identifying duplicates in different collections.

Descriptor and Descriptor States

Qualitative Descriptor

1. Cotyledons leaf colour: 1= Light green, 2= Green, 3= Dark Green
2. Cotyledon size: 1= Small, 2= Medium, 3= large
3 Stem pubescence density: 0= Absent, 3= Sparse, 5= Medium, 7= Dense
4. Stem shape: 1=Rounded, 2= Angular, 99= Others
5. Tendril: 0= Absent, 1= Present
6. Tendril type: 1= Coiled, 2= Straight, 99=others
7. Tendril branching: 1= Unbranched, 2= Branched, 99=Others
8. Leaf pubescence: 0= Absent, 1= Present
9. Leaf pattern: 1= Normal, 2 = Variegated, 99=Others
10. Leaf shape:1= Ovate, 2= Oblong, 3 = Cordate, 4 = Orbicular, 99=Others
11. Leaf size: 3 = Small, 5 = Medium, 7 = Large
12. Leaf margin: 1 = Entire, 2 = Serrate, 3= Multi-fid
13. Flower colour: 1 = white, 2= Creamy, 3= Yellow, 99= Others
14. Fruit shape: 1 = Spindle, 2= Elongated spindle, 3= Lengthened cylindrical, 99= Others
15. Fruit curvature: 1= Straight, 2= Medium curved, 3=Highly curved
16. Fruit skin primary colour:1= Light green, 2= Green, 3= Dark green, 99= Others
17. Fruit skin secondary colour: 1= Green with splashes, 2 = Dark green with splashes, 99= Others
18. Fruit skin lustre: 3=Matt/ Dull, 5 = Intermediate, 7 = Glossy

Quantitative Descriptor

1. Vine length (cm): On four random plants, data were collected from the ground level to the tip of the principal branch/vine and averaged at full fruiting (on marketable fruits).
2. Number of primary branches: Data were collected as an average of the same four plants at the end of the flowering stage.
3. Days to 50% flowering: Days required for 50% of the flowers to open
4. Days to first fruit harvest: Data was documented as the number of days between the date of sowing/ transplanting and the first marketable fruit harvest.
5. Fruit length (cm): Data were recorded as an average of 5 random fruits at the marketable stage.
6. Fruit width (cm): Data were recorded as the average of the same 5 random fruits at the marketable stage.
7. The number of fruit per plant: Data were recorded at the fruiting stage till the last harvest.
8. Individual fruit weight (g): Data were noted as an average of 5 fruits per accession.
9. Fruit yield per plant (kg): Five fruits were randomly selected from each germplasm and their average weight was taken as the individual fruit weight
10. Number of seeds per fruit: Data were recorded after harvest average of 5 random mature fruits
11. 100 seed weight (g): Data were calculated as the average weight of 100 random dry seeds
12. Yield per plot (kg): Data were taken after the harvest average of four plants' total fruit yield.

RESULTS AND DISCUSSION

Qualitative Character

Qualitative traits play an important role in describing a plant's physical appearance (Khatun *et al.*, 2023). Many aspects are influenced by physically observed traits including the natural se-lection of plants, customer preferences, and the socioeconomic setup of an area (Ghafoor *et al.*, 2002). The morphological observations of 33 germplasm are presented in Table 2. Among the whole (17), eight germplasm showed distinct variations in cotyledonous leaf colour, cotyledon size, stem pubescence density, leaf margin, fruit shape, fruit curvature, fruit skin's primary col-our, fruit skin secondary colour, and fruit skin luster while the nine had no remarkable varia-tion. Ara *et al.* (2015) reported a similar result among the 34 snake gourd genotypes in terms of qualitative attributes such as fruit colour, fruit shape, lamina type, fruit stripe,

and skin texture. After germination, cotyledonous leaf colours were observed in light green (45.05%) and green colour (45.95%). Cotyledon size was found medium (54.54%) and small (45.45%). Stem pu-bescence density was determined to be sparse (42.42%), medium (42.42%), and dense (15.15%).

Leaf margin was serrate amongst (42.42%) followed by entre (36.36%) and multifid (21.21%). The fruit-shaped spindle appeared (30.30%) whereas an elongated spindle (24.24%), and lengthened cylindrical were found (12.12%). Ara *et al.* (2015) also found variations in three different types of fruit shapes of snake gourd viz. cylindrical, elongated, and sickly. Fruit curvature was classified as straight, medium curved, and highly curved. The fruit curvature maximum was observed straight type (72.73%), followed by the medium curved (27.27%), and highly curved (6.06%). Fruit colour is a key factor in determining consumer preference (Ah-med *et al.*, 2022). The fruit skin's primary colour exhibited light green, green, dark green, and whitish, 51.52%, 30.30%, 12.12%, and 6.06%, respectively. A similar finding was noted by Ara *et al.* (2015). Fruit skin secondary colour was recorded in the maximum green with splashes (84.85%) whereas dark green with splashes (9.06%) and whitish with splashes secondary colour was presented in the minimum germplasm (6.06%).

Table 2: Qualitative variation of different characters in snake gourd germplasm

Characters	Descriptor state	Number of germplasm	Percent of germplasm
Cotyledons leaf colour	Light green	15	45.45
	Green	18	54.54
Cotyledon size	Small	15	45.45
	Medium	18	54.54
Stem pubescence density	Sparse	14	42.42
	Medium	14	42.42
	Dense	5	15.15
Stem shape	Angular	33	100
Tendril	Present	33	100
Tendril type	Coiled	33	100
Tendril branching	Branched	33	100
Leaf pubescence	Present	33	100
Leaf pattern	Normal	33	100
Leaf shape	Reniform	33	100
Leaf margin	Entire	12	36.36
	Serrate	14	42.42
	Multifid	7	21.21
Flower colour	White	33	100
Fruit shape	Spindle	10	30.30
	Elongated spindle	8	24.24
	Lengthened cylindrical	5	15.15
Fruit Curvature	Straight	24	72.73
	Medium curved	7	27.27
	Highly curved	2	6.06
Fruit skin's primary colour	Light green	10	30.30
	Green	17	51.52
	Dark green	4	12.12
	Whitish	2	6.06
Fruit skin secondary colour	Green with splashes	28	84.85
	Dark green with splashes	3	9.09
	Whitish with splashes	2	6.06
Fruit skin luster	Matt/Dull	4	12.12
	Intermediate	17	51.52
	Glossy	12	36.36

Quantitative Character

Quantitative characters are important indicators that show high variability (Iqbal, 2015). The range, mean, standard deviation, and coefficient of variation of twelve quantitative characters of snake gourd have been presented in Table 3. The vine length varied from 410 to 195 cm. The maximum vine length (410cm) was in AR-261 which was followed by SA-32 (390 cm) and AC-423 (388cm) while the mini-mum vine length was obtained in AC-187 (195cm). In terms of the number of primary branches, there were significant differences between genotypes. It varied between 4.33 (AHI-99) and 15 (AC-46). Days to 50% flowering trait showed considerable variation among genotypes. The maximum number of days required to reach 50% flowering was estimated to be 88. AC-423 had the earliest flower blooming (43 days). Rahman *et al.* (1990) found similar observations of flowering differences in various cucurbits such as ridge gourd, bitter gourd, and bottle gourd. Mendligner *et al.* (1992) also stated that they as-sessed some of the collected pumpkin accessions and found that the days to bloom ranged from 57.3 to 88.3 days. The days to first fruit harvest range was 64 to 88 days and the average first harvest date was (72.2 days). Fruit length, fruit breadth, and fruit weight are key quantitative features that contribute considerably to production (Ahmed *et al.*, 2004). The Fruit length ranged from 13.3 to 51.2 cm. The longest fruit length was obtained in genotype AHI-104 (51.2 cm) followed by R-1 (47.4cm) while the shortest fruit length was observed in IAH-194 (13.3cm). Maximum fruit Width was found from the genotype SA-32 (5.9 cm) whereas the minimum was found in I-4 (3.53 cm). The highest number of fruits per plant was obtained in AHI-104 (15), while the lowest number of fruits per plant was identified in AHI-118 (3). These findings are corroborated by Haque *et al.* (2014). The weight of the fruit varied enormously amongst genotypes, the germplasm AR-239 (292g) scoring best in terms of individual fruit weight. The highest fruit yield per plant was obtained in AHI-104 (3 kg/plant), which was statistically equal to AR-39 (2.92 kg/plant), while the lowest was recorded in LAH-294 (0.32 kg/plant). Variations in yield per plant were also observed in watermelon (Chezhiyan, 1984), bottle gourd (Sharma *et al.*, 2013), musk melon (Swamy *et al.*, 1984), and pumpkin (Rana *et al.*, 1986; Shah *et al.*, 1992).

Number of seeds per fruit, 100 seed weight, yield per plot ranged from 11-64, 24-36g, and 1.28-12kg, respectively. The coefficient of variation (CV%) compares the rate of variation among crop plant attrib-utes (Sharma, 1988). The yield plot-1 (49.44%) achieved the highest CV% which was followed by the fruit yield per plant (49.22%), the number of primary branches (38.92%), and the number of fruits per plant (39.37%), and the number of seeds per fruit (37.16%). On the other hand, the lowest CV% was recorded in 100 seed weight (10.61%) which was followed by days to first fruit harvest (11.40%). These findings demonstrate that among the analyzed snake gourd traits, the yield per plot, fruit yield per plant, number of primary branches, number of fruits per plant, and number of seeds per fruit had the highest level of exploitable genetic variability.

Table 3: Quantitative variations of different characters in snake gourd germplasm

Character	Range		Mean	Standard deviation	Coefficient of variation %
	Min	Max			
Vine length (cm)	195	410	327.6	49.01	14.96
Number of primary branches	4.33	15	7.48	2.91	38.92
Days to 50% flowering	43	87	54.0	10.11	18.77
Days to first fruit harvest	64	88	72.2	8.23	11.40
Fruit length (cm)	13.3	51.2	32.0	8.55	26.40
Fruit width (cm)	3.53	5.9	4.66	0.66	14.12
Number of fruits per plant	3	15	7.12	2.80	39.37
Individual fruit weight (cm)	70	192	182	60.41	33.19
Fruit yield per plant (kg)	0.32	3	1.28	0.63	49.22
Number of seeds per fruit	11	68	32.5	12.08	37.16
100 seed weight (g)	24	36	29.3	3.11	10.61
Yield per plot (kg)	1.28	12	5.13	2.45	49.44

Multivariate Analysis

Multivariate approaches are well-established tools for estimating variability and correlations among germplasm. Multivariate analytic approaches such as principal component analysis and cluster analysis were used to investigate the link between the 33 snake gourd germplasm and various morphological quantitative features and to classify the population.

Principal Component Analysis (PCA)

The principal component analysis is a technique for identifying plant features that account for the majority of the observed variance among genotypes. This tool is useful in the selection of the best genotypes for

breeding purposes (Ulaganathan & Nirmalakumari, 2015). Table 4 and Figure 1 display the PCA results of yield and yield contributing quantitative traits of snake gourd germplasm. The percentage of variation associated with each principal component was obtained by drawing a graph between eigenvalues and principal component numbers through a scree plot. The eigenvalue of the first principal component accounted for 30.6% of the total variation. The eigenvalues gradually declined from PC1 to PC12. The Eigenvalues for left out principal components were 18.3, 15.9, 10.5, 9.3, 5.2, 4.6, 2.5, 1.8, and 1.3% of total variation presented among the gen-otypes, respectively (figure 1). The first five PC axes explained 84.52% of the variation, suggesting con-siderable diversity among the genotypes for all the characters, the rest of the components were not con-sidered. Eigenvalues greater than 1.0 suggested that the detected features within these axes had a sig-nificant influence on the phenotype of the cultivar and could be effectively employed for selection among them. According to (Jahan *et al*., 2022), the first two components out of twelve traits had ei-genvalues up to 1.0, resulting in a cumulative variance of 84.10%. The PC1 had a higher positive con-tribution than the others. The traits, namely the yield per plot (0.916), fruit yield per plant (0.910), in-dividual fruit weight (0.683), fruit length (0.635), number of fruits plant^{-1} (0.562), fruit width (0.425) and number of seeds fruit^{-1} (0.396) are crucial for enhancing yield and quality traits. The PC2 had high positive contribution to number of seeds per fruit (0.789), number of primary branches (0.724), fruit width (0.499), days to first fruit harvest (0.387) and fruit length (0.351). The PC3 had maximum posi-tive contribution in days to first fruit harvest (0.796) and days to 50% flowering (0.779). Moreover, the PC4 had maximum positive contribution in vine length (0.556) and individual fruit weight (0.406). On-ly vine length (0.397) was the maximum contribution of PC5. Therefore, these traits can be chosen for a hybridization program to develop elite lines or F1 hybrids. Many scholars, including Kumar, 2015 and Pradhan *et al*., 2018 highlighted the comparable results.

Table 4: The eigenvalues and contributions of several characteristics of snake gourd germplasm to the major principal component

Characters	PC1	PC2	PC3	PC4	PC5
Vine length (cm)	-0.321	0.031	-0.484	0.556	0.397
Number of primary branches	0.099	0.724	-0.094	-0.271	0.282
Days to 50% flowering	-0.431	0.066	0.779	0.309	0.151
Days to first fruit harvest	-0.101	0.387	0.796	0.261	0.147
Fruit length (cm)	0.635	0.351	-0.045	-0.375	0.246
Fruit width (cm)	0.425	0.499	0.120	0.451	-0.343
Number of fruits plant^{-1}	0.562	-0.550	0.438	-0.165	0.266
Individual fruit weight (g)	0.683	0.211	-0.273	0.406	-0.389
Fruit yield per plant (kg)	0.910	-0.312	0.146	0.092	0.005
Number of seeds per fruit	0.396	0.779	0.088	-0.199	0.045
Seed weight (g)	-0.393	0.002	0.293	-0.348	-0.656
Yield per plot (kg)	0.916	-0.312	0.146	0.092	0.005
Eigenvalue	3.67	2.20	1.90	1.26	1.11
Variation (%)	30.55	18.33	15.86	10.50	9.28
Cumulative variance (%)	30.55	48.88	64.74	75.24	84.52

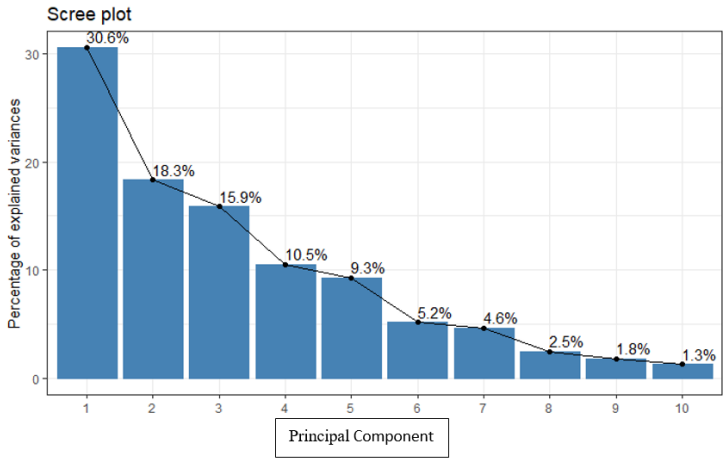

Figure 1: Contribution on variation percentage among the germplasm based on principal component analysis

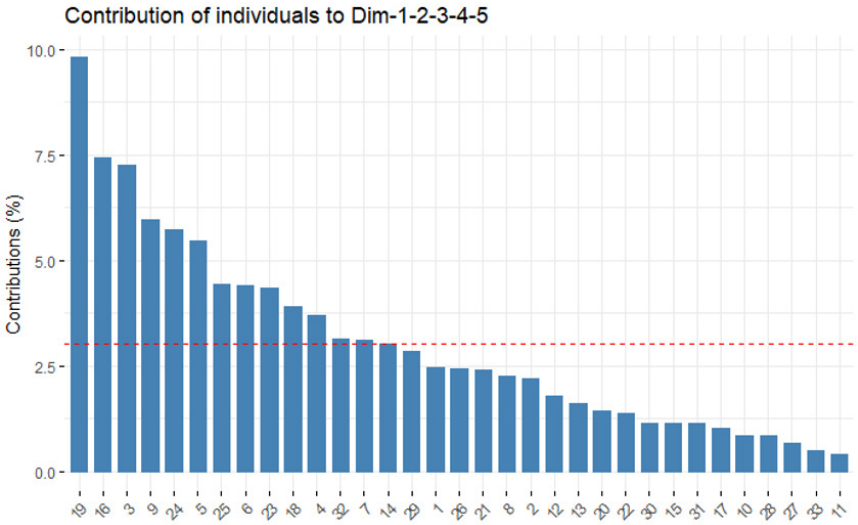

Figure 2: Contributions of individual germplasm among the 33 germplasm

Figure 2 depicts the contribution of each germplasm among the 33 genotypes. The germplasm 19 (AHI-99) made the most contribution. The second greatest contributing germplasm was discovered at 16 (IAH-68), 3 (N-170). Furthermore, 9 (AC-423), 24 (R-1), and 5 (AC-46) contributed moderately. The lowest contribution was recorded in 11 (AR-48), followed by 33 (RISA-126), 27 (SA-32), 28 (KASI-34), 10 (RA-39), 17 (IAH-294), 31 (RIM-50), 15 (RA-276), and 30 (ATR-37).

PCA Biplot Analysis

A two-way form of genotype-traits biplot displayed the comparative value of the traits which was being prepared from the 12 parameters and 33 genotypes (Figure 3). Biplot analysis is mostly used to deter-mine the components whose effect is more to create the genotypic variation. The highest values indi-cate the highest influence of the trait on the total variation. Biplot analysis determines varietal stability in the multi-environmental trial (Farshadfar *et al.*, 2013). It describes the association among the traits across different genotypes (Yan and Reid, 2018). In addition, biplot analysis showed the trait profiles of the genotypes, especially, those genotypes positioned far away from the results indicated a correlation between traits with genotypes. GT biplot, a vector drawn from the origin of each trait facilitates the visualization of interrelationships among traits. The vector length of the trait measures the magnitude of its effects on the yield view of the GT biplot is best to visualize patterns between genotypes and traits, provided the biplot should explain a sufficient amount of the total variation. The correlation coef-ficient between any 2 traits is approximated by the cosine of the angle between their vectors (Yan and Rajcan, 2002). The first PC explained 30.6% of the total variability and the second PC explained 18.3% of the variation among 33 snake gourd germplasm. Thus, the present study revealed that the first PC was more important than the second PC for explaining the variability among the germplasm based on studied traits. Begum *et al.* (2013) observed that the first axes accounted for 20.07% variation among the genotype. The germplasm N-170, LAH-68, AHI-99, R-1, KI-36, AC-423, AC-187, and AC-118 were separately isolated from the others and they were away from the centroid. This result showed the uniqueness and divergence of the germplasm. Similar results have been reported by many researchers, such as Kumar *et al.*, (2014) for cucumbers; Geetha and Divya, (2021) for sorghum, and Reddy *et al*, (2015) for foxtail millet.

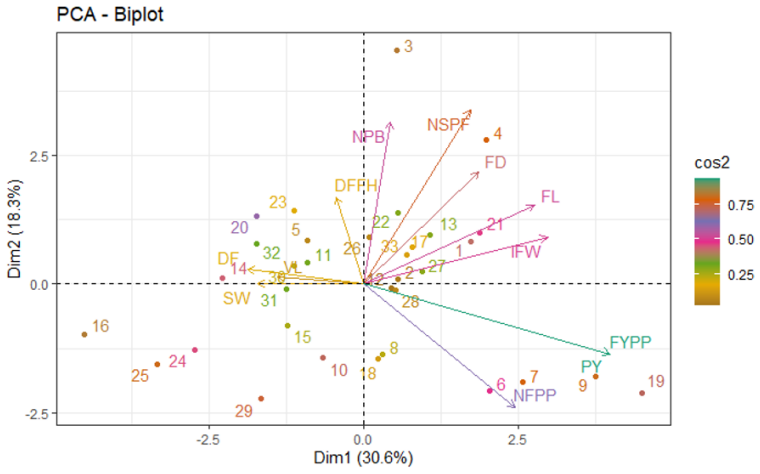

Figure 3: A G×T biplot based on germplasm 33 and 12 morpho-physiological traits of germplasm

Cluster Analysis

Cluster analysis is a method for categorizing the materials into groups. It is a reliable technique to de-termine the similarities and extent of differences within various germplasm (White *et al.*, 2012). It could be useful for future breeding programs. According to the cluster analysis, five groups of germplasm were identified under groups I, II, III, IV, and V including the number of germplasm 4, 10, 3, 5, and 11, respectively, In addition, the dendrogram was used to show the clustering pattern (Table 5). Cluster V was discovered as the largest group followed by cluster II while cluster III consisted minimum number of germplasm (3) followed by cluster I (4) and IV (5). A similar finding was recorded in pumpkin Chaudhari *et al.* (2017); Kalyani Pradhan *et al.* (2018) in ash gourd, and Premchand *et al.* (2017) in ridge gourd.

Table 5: Distribution of 33 snake gourd germplasm into five distinct clusters

Cluster	Number of Germplasm	Constituent germplasm
I	4	AC-21, N-102, AC-46, TRMR-130
II	10	N-170, MAH-45, RAI-125, MAH-03, AC-423, AR-261, SA-32, AR-262, KASI-34, KI-36
III	3	AC-118, AR-276, R-69
IV	5	AR-39, AHI-104, AHI-99, AC-187, AC-357
V	11	IAH-68, AR-48, ATR-37, IAH-294, I-4, R-1, RISA-126, AR-188, AHI-118, RIM-50, MK-90

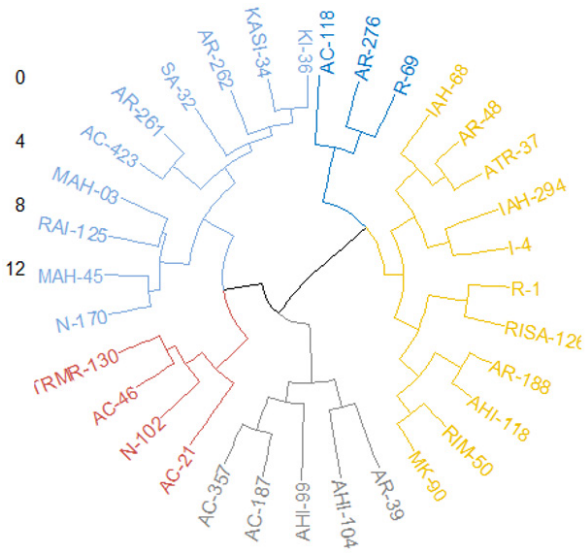

Figure 4: Dendrogram of snake gourd germplasm based on quantitative characters obtained by cluster analysis where the color blue, yellow, ash, red, and sky blue represent the clusters III, V, IV, I, and II, respectively

Mean Value of Quantitative Parameters by Cluster Analysis

Cluster means to represent the genetic variations between clusters. The mean value for twelve charac-ters concerning five clusters is presented in Table 6. Variations in cluster mean were identified for all characters. Of the five clusters, the maximum cluster mean value was observed in cluster IV for the character's fruit length (36.44 cm), number of fruits plant^{-1} (11.60), fruit yield per plant (2.34 kg), and yield per plot (9.37 kg). Gautam *et al.* (2017) reported that clustering revealed instability due to considerably less divergence, but significantly diverse clusters remain distinct in varied contexts. Cluster I ex-hibited the highest mean value for the number of primary branches (12.13), fruit width (5.35 cm), and number of seeds per fruit (48.75). Similarly, cluster III also found the three highest mean values vine length (346.67 cm), days to 50% flowering (80.33), and days to first fruit harvest (84.67). In case of Cluster II and cluster V had the maximum mean value in individual fruit weight (233 g) and seed weight (30.27 g), respectively. The rest of the traits showed moderate value. Sing *et al.* (2013), Chaudhari, (2017), and Kalyani Pradhan, (2018) obtained similar results in bitter gourd, pumpkin, and ash gourd, respectively. Based on the cluster mean, the important cluster is cluster VI which had the highest mean values for fruit length, number of fruits plant-1, fruit yield per plant, and yield per plot. Through the hybridization approach, this group of accessions may be preferred for transferring desirable characteristics. For a good bleeding of genetic potential, genotypes should be selected based on cluster mean (Wolie *et al.*, 2013).

Table 6: Cluster mean for twelve characters of snake gourd germplasm

Characters	Cluster I	Cluster II	Cluster III	Cluster IV	Cluster V
Vine length (cm)	320.50	343.80	346.67	264.20	339.18
Number of primary branches	12.13	7.36	6.33	5.17	7.26
Days to 50% flowering	56.75	47.50	80.33	50.40	53.00
Days to first fruit harvest	83.25	68.40	84.67	70.00	69.27
Fruit length (cm)	36.13	33.96	24.13	36.44	28.39
Fruit width (cm)	5.35	4.99	4.80	4.50	4.13
Number of fruits plant^{-1}	7.50	6.00	7.67	11.60	5.82

Individual fruit weight (g)	207.00	233.00	133.67	204.40	129.55
Fruit yield per plant (kg)	1.55	1.37	0.91	2.34	0.73
Number of seeds per fruit	48.75	34.57	24.33	29.14	28.52
Seed weight (g)	29.50	28.40	29.33	28.60	30.27
Yield per plot (kg)	6.19	5.47	3.63	9.37	2.93

CONCLUSION

The studied snake gourd germplasm showed a wide range of variations. The maximum variation was recorded in fruit shape, fruit curvature, and fruit skin's primary colour and fruit skin's secondary colour. According to the principal components analysis, the first two (PC) contributed the maximum variabil-ity. Similarly, in cluster analysis, cluster IV showed a higher mean value by more characters that could be effectively used for the breeding program. In terms of quantitive characters, the fruit length, fruit width, fruit weight, number of fruits per plant, and days to first fruit harvest were significant contribu-tors to the variability. Of the snake gourd germplasm N-170, LAH-68, AHI-99, R-1, KI-36, AC-423, AC-187, AC-118, AR-39, AHI-104, AHI-99, and AC-357 performed better which can be selected as promising germplasm for breeding programs.

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