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Morpho-Diversification Study on Cluster Bean [*Cyamopsis tetragonoloba* L. (Taub)] Using Distinctiveness (D), Uniformity (U) and Stability (S) Descriptors

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ABSTRACT

The present investigation was conducted in the month of January to April, 2022 in Poothurai village of Villupuram district to assess, evaluate and identify the morphological variations in cluster bean accessions. One hundred and two genotypes, collected from different geographic locations of the country were utilised for the study. The cataloguing of qualitative and quantitative descriptors was done based on the Distinctness, Uniformity and Stability standards provided by PPV & FRA, 2001. Twenty-two descriptors with sub-groups were taken for the study. The qualitative trait includes; plant type, leaf pubescence, leaf serration, leaf shape, petiole length, terminal leaflet length, leaf color, flower color, immature pod color, seed shape and seed coat color. The quantitative trait comprises of plant height, number of primary branches, days to maturity, days to 50 % flowering, number of clusters per plant, number of pods per cluster, pod length, number of pods per plant, number of seeds per pod, seed size and gum content. The genotypes were scored according to their characteristic feature in each of the sub-groups and their frequency of occurrence was recorded. The present study clearly indicates the utilization of DUS standards and their importance in conservation, maintenance and protection of accessions in order to ensure their use in development of novel varieties.

Keywords Cluster bean, DUS, Descriptors, Qualitative, Quantitative.

INTRODUCTION

Cluster bean (*Cyamopsis tetragonoloba* (L.) Taub) is a versatile crop belonging to the arid and semi-arid regions of India and Pakistan (Mahla *et al*. 2020). Belonging to the Fabaceae family this drought-resilient crop can be highly cultivated on the marginal areas of dry lands (Kumar *et al*. 2017). It is a multi-purpose crop that can be utilised as a vegetable, fodder for feed and guar gum which have immense industrial potential especially as an export commodity to major

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developed countries. India, being the largest producer of the crop, contributes to 80 % of total world production. Owning to its importance in industrial sector its cultivation has been wide-spread to the non-traditional regions. Cluster bean is a cheap source of protein and other nutrients (Bhatt *et al*. 2016). Apart from being known to fix atmospheric nitrogen, it adds organic carbon to the soil by shedding its leaves gradually up until maturity thereby rejuvenating soil health (Jukanti *et al.* 2015).

Having originated from the South East Asian countries, cluster bean has had wide variability and diversity in this specific geographic location. In order to exploit this diversity to maximum potential, it is essential to distinguish the germplasm and categorize the descriptors so that they can be utilised as genetic markers (Yadav 2021). For management, maintenance and utilization of Plant Genetic Resources (PGR) for crop cultivar development, it is indispensable to use well defined descriptors. Characterization and assessment help to increase the utilisation of germplasm by providing better insights into the genetic diversity and composition of the collection (Byregowda *et al.* 2015). DUS listing helps in identifying a novel variety from existing accessions with consistently expressed characters that do not change over generations as qualitative characters are pronounced to be more stable in nature (Raut 2003). The introduction of Protection of Plant Varieties and Farmers Rights (PPV&FR) act in 2001, acts as a platform for the criterion required for DUS testing. Thus, this study aims at establishing an elaborate morphological based DUS characterisation of cluster bean accessions collected from various locations of the country.

MATERIALS AND METHODS

Experimental Site

To analyse and screen the cluster bean germplasms, a detailed investigation was performed from January to April 2022 in a field in Poothurai Village, Villupuram District, Tamil Nadu. The experimental site is set 23 metres above mean sea level at 11.9667° North Latitude and 79.7596° East Longitude. With an average summer temperature of 32 °C and an average winter temperature of 24.6 °C, the annual average temperature is 28.4 °C. Red soil with good water-holding capacity can be found in the experimental area.

Table 1 List of genotypes collected from various regions.

Sl. No	Number of Genotypes	Source
1	50	ICAR -National Bureau of Plant Genetic Resources, Regional Station, Jodhpur
2	30	Private sectors
3	8	Released varieties
4	14	Local collection

Materials and field plot

A total of 102 genotypes (including the check variety Pusa Navbahar) from various geographic regions of the country were used (Table 1). These were assessed using Randomised Block Design (RBD), with ridges and furrows spaced 45 cm between ridges and 15 cm between plants. Five metre long single rows were taken for sowing the seeds of the genotypes. Throughout the whole cropping season, optimal management techniques advocated by the Tamil Nadu Agricultural University was implemented.

Observations recorded

DUS cataloguing of cluster bean genotypes were conducted as per the standards provide by Protection of Plant Varieties and Farmers' Rights Authority (PPV & FR). Ten random plants were selected for every genotype, the data was recorded for both qualitative and quantitative traits. For the qualitative traits, plant type was recorded according to its framework. Leaf pubescence, leaf shape, leaf serration, leaf color, leaf petiole length and terminal leaflet length was recorded on 5th fully opened leaf from base on main stem. The color for flowers, leaves, pod and seed coat were recorded in accordance with those provided by Royal Horticultural Society. Foliage colour was scored as light green (RHS 135D), medium green (RHS 130 A) and dark green (RHS 134 A). Flowers varied with colors like white (RHS 155A) and Purple (RHS 075B). The flowers with higher 50 % of inflorescence at early morning was recorded. The color of immature pods was scored as green (RHS 130A) and Dark green (RHS 134A). The seed coat color was

also classified as; light yellow (159 C), Yellow brown (163 C), Grey (N 200D) and Black (N202A). The seed shape was classified as semi-flat and globular. The quantitative characters recorded include, plant height (cm), number of primary branches, days to maturity, days to 50 % flowering, number of clusters per plant, number of pods per cluster, pod length (cm), number of pods per plant, number of seeds per pod, seed size and gum content. All the parameters were scored individually, the number of genotypes with similar characters were recorded and their frequency of occurrence was expressed in percentage.

RESULTS AND DISCUSSION

To facilitate genotype use and also to distinguish a candidate variety from all other categories, adequate morphological and agronomic trait differentiation is necessary for affording the plant breeder the rights to protect within the system of intellectual property rights. The recording of distinctly evident, heritable traits is known as genotype characterization. Genetic resources can be identified by a variety of features, especially morphological, agronomic, biochemical and molecular (Umesha *et al*. 2015). Being an affordable and time saving method, preliminary characterisation of genotypes is preferred for identifying the variations in morphological, agronomic and qualitative traits of the accessions. The results are presented in (table 2).

Growth characters

Based on plant height, the accessions were categorized into three groups of which the major group was tall types with 86.27 % of occurrence followed by medium (13.73%). None of the accessions fell under the short group. Three groups were registered for plant type of which 22.55 % were erect, 47.06 % were semi-erect and 30.39 % were spreading in nature. In case of number of primary branches, 68 accessions fell under the low group having less than 5 primary branches, whereas 22 and 12 genotypes were observed to have medium (5 - 8) and high (> 8) number of primary branches respectively. They contributed to 66.67 %, 21.57 % and 11.76 % frequency of occurrence respectively. In accordance with days to maturity, most of the genotypes fall under medium (44.12 %) and late (33.33 %) maturing types. Only about 19.61 % were

Table 2. Frequency of distribution and scoring of cluster bean accessions to various DUS descriptors.

Sl. No	Characteristics	Reference Character	Score	Number of genotypes	Frequency of occurrence (%)
Growth characters					
1	Plant height	Short (< 50)	3	0	0.00
		Medium (50 - 70)	5	14	13.73
		Tall (> 75)	7	88	86.27
2	Plant type	Erect	3	23	22.55
		Semi-erect	5	48	47.06
		Spreading	7	31	30.39
3	Number of primary branches	Low (< 5)	3	68	66.67
		Medium (5 - 8)	5	22	21.57
		High (> 8)	7	12	11.76
4	Days to maturity	Early (< 90)	3	20	19.61
		Medium (90 - 110)	5	45	44.12
		Late (> 110)	7	34	33.33
5	Leaf pubescence	Absent	1	45	44.12
		Present	9	57	55.88
6	Leaf serration	Absent	1	44	43.14
		Medium	3	15	14.71
		Strong	5	43	42.16
7	Leaf shape	Narrow	3	60	58.82
		Broad	5	42	41.18
8	Foliage colour	Light green	3	35	34.31
		Medium green	5	27	26.47
		Dark green	7	40	39.22
9	Terminal leaflet length	Short (< 4)	3	12	11.76
		Medium (4 - 6)	5	55	53.92
		Long (> 6)	7	35	34.31
10	Leaf petiole length	Short (< 2.5)	3	37	36.27
		Medium (2.5 – 3.5)	5	24	23.53

Table 2. Continued.

Sl. No	Characteristics	Reference Character	Score	Number of genotypes	Frequency of occurrence (%)
		Long (> 3.5)	7	41	40.20
Flower and pod attributes					
11	Days to 50% flowering	Early (< 45)	3	88	86.27
		Medium (45 - 55)	5	14	13.73
		Late (> 55)	7	0	0
12	Flower colour	White	3	15	14.70
		Purple	5	87	85.29
13	Number of clusters per plant	Low (< 12)	3	20	19.61
		Medium (12 - 20)	5	51	50
		High (> 20)	7	31	30.39
14	Number of pods per cluster	Low (< 5)	3	71	69.61
		Medium (5 -8)	5	24	23.53
		High (> 8)	7	7	6.86
15	Pod length (cm)	Short (< 10)	3	95	93.14
		Medium (10 - 15)	5	7	6.86
		Long (> 15)	7	0	0.00
16	Immature pod colour	Green	3	72	70.59
		Dark green	5	30	29.41
17	Number of pods per plant	Low (< 60)	3	39	38.24
		Medium (60 - 80)	5	23	22.55
		High (> 80)	7	40	39.22

Table 2. Continued.

Sl. No	Characteristics	Reference Character	Score	Number of genotypes	Frequency of occurrence (%)
Seed parameters					
18	Number of seeds per pod	Low (< 6)	3	22	21.57
		Medium (6 - 8)	5	70	68.63
		High (> 8)	7	10	9.80
19	Seed shape	Semi-flat	3	80	78.43
		Globular	5	42	41.18
20	Seed size (weight of 100 seeds)	Small (< 2.5)	3	10	9.80
		Medium (2.5 – 3.5)	5	89	87.25
		Large (> 3.5)	7	3	2.94
21	Seed coat colour	Light yellow brown	3	30	29.41
		Yellow brown	5	37	36.27
		Grey	7	25	24.50
		Black	9	10	9.80
22	Gum content (%)	Low (< 30)	3	95	93.14
		High (> 30)	5	7	6.86

grouped under early maturing types with a growing period of less than 90 days. Among the qualitative traits, bushy growth habit was observed in a dominant spectrum over single-stemed types. Single-stemed genotypes, because of their ability to bear higher number of pods or clusters are often preferred compared to the spreading types, provided, they can also be effectively utilised as an intercrop (Kumara et al. 2017). Earlier maturing varieties are usually preferred for any crop improvement programme, enabling multiple crop seasons in a single year.

In all the genotypes studied, 55.58 % of the accessions had pubescent leaves and stems and the remaining 44.12 % were glabrous. Three groups were identified for leaf serration *viz*, absent, medium

and strong, of which 43.14 % (44 genotypes) had no serrations, 14.71 % (15 genotypes) and 42.16 % (43 genotypes) were medium and strongly serrated respectively. Most of the genotypes fell under narrow shaped leaves with 58.82 % of occurrence and the remaining 41. 18 % were broad leaved. Pubescent, serrated and narrow leaves were predominantly observed among the genotypes. The leaf type and surface might be employed to distinguish between gum and vegetable genotypes. The distinctive characteristic of a broad and glabrous leaf surface aids in categorizing a genotype as vegetable. However, gum types are linked to narrow leaves with pubescence. These parameters were utilized by Kumar *et al*. (2013) to characterize the guar variety. Even at the earliest stages of crop growth, such specific morphological characters can be employed in direct separation of vegetable genotypes as off-types from the cluster bean seed production plots.

Foliage color was ranked as green, medium green and dark green with frequency percentage of 34.31 %, 26.47 % and 39. 22 % respectively. The terminal leaflet length was categorized into three groups such as short (< 6), medium (4 - 6) and long (> 6) of which 12 were short (11.76 %), 55 were of medium (53.92 %) length and 35 were longer (34.31 %). The length of the petiole had three groups of which, 36.27 % (37 genotypes) had a shorter petiole of less than 2.5 cm, 23.53 % (24 accessions) had medium petioles (2.5 – 3.5) and the remaining 41 genotypes had longer petioles of more than 3.5 cm with a percentage occurrence of 40.20 % (Table 2, Fig. 1 A, B, C, D).

Flower and pod attributes

Days to 50 % flowering varied for the different accessions and were grouped as early with 86.27 % of genotypes and medium with 13.73 % of genotypes. None of the genotypes were late flowering. This variations in flowering may also be attributed to the changes in environmental conditions. Two groups of flowers were recorded i.e., white and purple group (Table 2, Fig. 1E). About 85.29 % (87 genotypes) contributed to purple group, whereas, 14.70 % around 15 genotypes were grouped under white flowers. Purple colored flowers apparently appeared in most of the released varieties. Though, they do not have any specific reasons, studies suggest that flower color provides extra fitness under arid and semi-arid regions (Strauss *et al.* 2004). Also, Kumar *et al*. (2013) reported that flower color helps in reducing contamination during seed production thereby maintaining purity. Immature pods were grouped into two colors namely green and dark green. 70.59 % of the genotypes had green colored pods and 29.41 % of genotypes were dark green in color. The number of clusters per plant were low in 20 genotypes, medium in 51 accessions and high in 31 genotypes with an occurrence rate of 19.16 %, 50 % and 30.39 % respectively. Observations on number of pods per cluster revealed that 69.61 % of genotypes had pods less than 5 in a cluster, flowed by 23.53 % in medium (5 - 8) and 6.86 % in high (> 8) (Fig. 1 F and G).

Maximum number of genotypes are found to have short pods with an occurrence of 93.14 % and the remaining genotypes were medium in length. With respect to number of pods per plant, three groups were obtained; low (38.24 %) with 39 genotypes, medium (22.55 %) with 23 accessions and high (39.22 %) with 40 genotypes. Pod characters being quantitative in nature is highly variable. This variability in cluster number, pod length, pods per cluster and pods per plant, indicates a possibility of direct selection and evaluation of superior stocks over time and space (Umesha *et al*. 2015).

Seed characters

The number of seeds per pod were classified into three groups i.e., low (< 6), medium (6 - 8) and high (> 8), where the occurrence percentage of genotypes were 21. 57 %, 68.63 % and 9.80 % respectively (Fig. 1 I). Maximum number of genotypes (80) were falling under semi-flat shaped seeds and the remaining 42 were globular in shape (Fig. 1 H). About 9.80 % of accessions were small seeded (< 2.5 g), 87.25 % were medium sized (2.5 – 3.5 g) and the remaining 2.74 % were large (> 3.5 g) seeded. The seed coat color was classified into four major groups (Fig. 1 J, K, L and M) with 29.14 % being light yellow brown, 36.27 % being yellow brown, 24.50 % being grey and 9.80 % of seeds owning black seed coat. The guar content was low (< 30) in 75 genotypes contributing to about 93.14 % and high (> 30) in 7 genotypes with

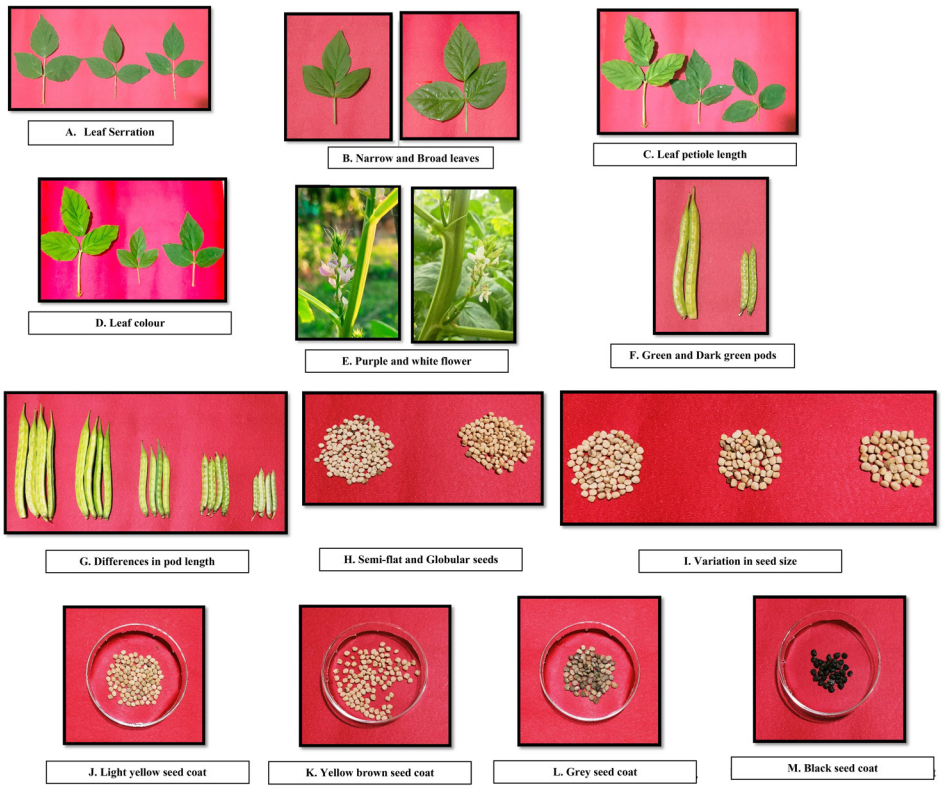

Fig. 1. The variations observed for the DUS characterisation is given below.

about 6.86 %. These variations in seed parameters can be predominantly utilized as a tool for selection of a variety when taken into consideration for seed production to exploit guar gum in industrial sectors. Similar findings on seed coat colour were also reported by Kumara *et al*. (2017), Sultan *et al*. (2012) and Morris (2010).

CONCLUSION

A wide range of variation was recorded during DUS characterisation of cluster bean accessions. For determining any pre-breeding programme, knowledge on both quantitative and qualitative characterisation is critical for germplasm conservation, development of novel varieties and selection of superior accessions. The study on morpho-diversification helps in broadening the genetic base indicating the variations existing in collected varieties. This study thereby indicates the importance of cataloguing and characterisation of accessions using DUS descriptors for the maintenance and protection of genotypes.

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