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Research article

Agro-morphological characterization of pigeonpea (*Cajanus cajan* (L.) Millsp.): Basis to breeding

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Abstract

Phenotypic characterization of crop genetic resources generates important information for plant breeders. The genetic diversity of Tanzanian pigeonpea germplasm was assessed using qualitative and quantitative traits and the best and complementary parents were identified for use in a pigeonpea breeding program. In total, 48 entries collected from Tanzania and Kenya were evaluated for 10 qualitative and 16 quantitative agro-morphological traits. Cluster analysis was used to separate genotypes into different clusters. The results from the combined analysis of variance showed that genotypes varied significantly for most of the traits studied. The overall Shannon-Weaver diversity index ranged from 0.10 (pod form) to 1.04 (pod color). Pod form, flowering pattern and base seed color showed little variation among genotypes. Based on principal component analysis, the traits accounting for most of the variation in the characterized pigeonpea collection were: days to 50% flowering, days to maturity, number of pods per plant, number of seed per plant, grain yield, leaf width and leaf area. The cluster analysis delineated the genotypes into three clusters. The most desirable genotypes with distinct attributes useful in future pigeonpea breeding were: Bangili, TZA 5463, Babati White, ICEAP 00040, ICEAP 00932, ICEAP 00557 and ICEAP 00554.

Introduction

Pigeonpea, *Cajanus cajan* (L.) Millsp. is an important crop for millions of people living in dry regions of the world as it is a multipurpose crop that integrates crop and livestock production, thus contributing to food security (Ayenan, 2017). Africa, especially Eastern Africa, is considered as a secondary center of diversity for this crop due to the presence of wild relatives (Songok et al., 2010). The crop has gained popularity among the farming community in many parts of Tanzania and the area of production has increased in recent years. It is generally grown by smallholder farmers with low input under rainfed conditions (Khoury et al., 2015, Saxena et al., 2018; Zavinon et al., 2019). In Tanzania, over 80% of pigeonpea dry seed is sold to external markets and the remaining is consumed locally mainly as a green vegetable (Lo Monaco, 2006).

The pigeonpea plant provides variable sources of proteins for poor smallholder farmers in the semi-arid tropics and it can survive in dry environments (Damor et al., 2016; Khoiriyah et al., 2017). The search for diversity in a germplasm collection is a way of identifying desirable genes for future utilization in breeding, where the diversity in crop species usually depends on mutation, recombination, selection and genetic drift (Bhandari et al., 2017). Cultivated pigeonpea has low polymorphism (Odeny, 2007) so that pigeonpea breeders are left with no options other than utilizing the wild relatives from the secondary, tertiary and quarterly sources using appropriate gene transfer methods

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to increase polymorphism (Sameer-Kumar et al., 2019). According to Saxena et al. (2014), the highest level of polymorphism in wild relatives and landraces are in India. The wild relative of cultivated species is considered as important source of increasing the level of genetic diversity of pigeonpea (Yadav, 2018). The wild relatives of pigeonpea are known for superior traits such as a high protein content, cleistogamy, cytoplasmic male sterility, dwarfing habit and various biotic and abiotic stresses (Yadav, 2018).

Genetic diversity in a crop species can be studied using different methods including morphological and/or phenotypic, biochemical and molecular markers (Mehmood et al., 2008). Morphological characterization is considered as a traditional method because it is simple and inexpensive, without requiring special facilities or procedures (Mehmood et al., 2008). Morphological characterization provides an understanding of the crop species based on the phenotype under field conditions but is greatly affected by the environment (Abdi et al., 2002; Fufa et al., 2005). In the past, morphological traits, both qualitative and quantitative, have been successfully used to study genetic diversity in pigeonpea (Kallihal et al., 2016; Navneet et al., 2017; Zavinon et al., 2019). Information regarding genetic diversity for distantly related genotypes would assist breeders in choosing desirable parents for introgression. Therefore, the objective of this study was to assess genetic diversity based on qualitative and quantitative traits and to identify the best parents for different traits that could be used in a pigeonpea breeding program.

Materials and Methods

Plant material

The experimental materials consisted of 48 pigeonpea germplasm samples collected from Tanzania Agricultural Research Institute (TARI)-Tanzania and International Crops Research Institute for the Semi-Arid Tropics (ICRISAT)-Kenya. Of the 48 genotypes, 7 were released varieties in Tanzania encompassing superior agronomic traits and were used as donor parents for a national pigeonpea improvement program. A further 20 genotypes were landraces collected from the National Plant Genetic Resources Centre, while 9 genotypes were landraces grown by farmers in major pigeonpea growing areas collected from farmers and 12 genotypes were improved breeding lines obtained from ICRISAT-Nairobi. Table 1 summarizes the genotypes used and their country of origin.

Table 1 List of pigeonpea genotypes evaluated, place of collection and country of origin

Genotype	Status	Origin	Country	Genotype	Status	Origin	Country
ICEAP 00040	Released cultivar	TARI	Tanzania	ICEAP 01172/2	Breeding line	ICRISAT	Kenya
ICEAP 00936	Breeding line	ICRISAT	Kenya	ICEAP 01154/15	Breeding line	ICRISAT	Kenya
Babati White	Landraces	Farmers	Tanzania	ICEAP 00979/1	Breeding line	ICRISAT	Kenya
TZA 253	Landraces	NPGRC	Tanzania	ICEAP 01172/2	Breeding line	ICRISAT	Kenya
TZA 2466	landraces	NPGRC	Tanzania	ICEAP 01154/2	Breeding line	ICRISAT	Kenya
TZA 2785	Landraces	NPGRC	Tanzania	ICEAP 0673-1	Breeding line	ICRISAT	Kenya
TZA 197	landraces	NPGRC	Tanzania	ICEAP 00554	Released cultivar	TARI	Tanzania
ICEAP 00557	Released cultivar	TARI	Tanzania	Mthawanjuni	Landraces	Farmers	Tanzania
ICEAP O1179	Breeding line	ICRISAT	Kenya	TZA 2514	Landraces	NPGRC	Tanzania
Bangili	Landraces	Farmers	Tanzania	TZA 2464	Landraces	NPGRC	Tanzania
ICEAP 00540	Breeding line	ICRISAT	Kenya	TZA 5596	Landraces	NPGRC	Tanzania
Kondoa	Landraces	Farmers	Tanzania	TZA 5582	Landraces	NPGRC	Tanzania
TZA 2692	Landraces	NPGRC	Tanzania	Arumeru	landraces	Farmers	Tanzania
No. 40	Landraces	Farmers	Tanzania	TZA 5463	Landraces	NPGRC	Tanzania
ICEAP 00911	Breeding line	ICRISAT	Kenya	TZA 2496	Landraces	NPGRC	Tanzania
TZA 250	Landraces	NPGRC	Tanzania	Tumia	Released cultivar	TARI	Tanzania
TZA 5464	Landraces	NPGRC	Tanzania	ICEAP 00932	Released cultivar	TARI	Tanzania
Komboa	Released cultivar	TARI	Tanzania	Ilonga	Landraces	Farmers	Tanzania
TZA 5557	Landraces	NPGRC	Tanzania	ICEAP 00576-1	Breeding line	ICRISAT	Kenya
TZA 2509	Landraces	NPGRC	Tanzania	Hombolo	Landraces	Farmers	Tanzania
Kiteto	Landraces	Farmers	Tanzania	TZA 5541	Landraces	NPGRC	Tanzania
TZA 5555	Landraces	NPGRC	Tanzania	TZA 2456	Landraces	NPGRC	Tanzania
ICEAP 01147	Breeding line	ICRISAT	Kenya	TZA 2807	Landraces	NPGRC	Tanzania
TZA 2439	Landraces	NPGRC	Tanzania	ICEAP 00053	Released cultivar	TARI	Tanzania

TARI = Tanzania Agricultural Research Institute; ICRISAT = International Crops Research for Semi-Arid Tropics; NPGRC = National Plant Genetic Resources Centre, Arusha, Tanzania

Experimental sites

The evaluations were conducted at TARI stations located at Ilonga Kilosa-Morogoro and Hombolo-Dodoma. The relevant climatic and soil data detailed below for these sites were obtained from Budotela (1995) and Dolo et al. (2017). The soils at the TARI-Ilonga Centre are moderately fertile and well-drained characterized as loamy soils. The area has a semi-humid climate with an average annual rainfall of 800 mm. The early rain starts in November and ends in January, followed by heavy rainfall between March and May. The area experiences a long dry season from June to October and the average annual temperature is 25–30°C. The soils at the TARI-Hombolo Centre are characterized as reddish-brown sandy clay soil, classified as Ustic Torriorthents (Budotela, 1995). The area has a semi-arid climate with an average annual rainfall of 350–500 mm. The rainy season is between December and March and April to November is the dry season. Sunshine persists for almost 12 hr/d, with wind speed in the range between 1.0 m/s in February and 4 m/s in October (Hussein, 2010). The average annual daily air humidity is 65% and the temperature is in the range 25–32°C. The rainfall data for Hombolo and Ilonga sites in the 2015/2016 season are presented in Fig. 1.

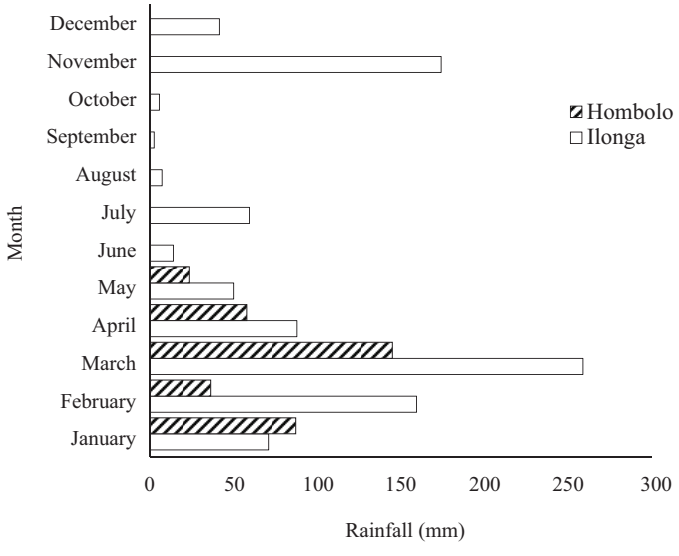

Fig. 1 Rainfall data for Hombolo and Ilonga sites in 2015/2016 season

Experimental layout, design and field management

The genotypes were evaluated for one rainy season at Ilonga during December 2015–August 2016 and at Hombolo during January–July 2016. The experiment was conducted using an 8 × 6 row-column design, with two replications at each site. The 48 genotypes were planted in 4-row plots, 3 m in length, with inter-row and intra-row spacing of 1 m and 0.5 m, respectively. Data were collected on the middle rows. To avoid a border effect, one border row was planted each side of the plot and the first and last plant in each row were excluded from the plot yield. Sowing of the experimental field was done after heavy rain to ensure even germination. Two seeds were sown per hill and, after germination, thinning was done to one plant per hill. The agronomic practices included an application of pre-emergence herbicide using 2, 4-dichlophenoacetic acid and Roundup (glyphosate) at a rate of 2–4 L/ha to control weeds. Three hand weeding sessions and three sprays of insecticides (Ninja 5 EC and Karate 5 EC at 30 ml/20 L) were applied to control pod borer and three sprays of fungicides (SUPER GRINO 76 WP at 2–2.5 kg/ha) were done to control fungal diseases. No fertilizer was applied to be consistent with common farmer practice.

Data collection and statistical analysis

The qualitative traits studied consisted of: growth habit, stem color, flower base color, pattern of streak, flowering pattern, pod color, seed eye color, pod hairiness, pod form, seed eye width, base seed color, seed shape, seed color pattern, leaf shape and leaf hairiness. The traits were evaluated based on the set standards for characters by the International Board of Plant Genetic Resources Institute descriptors for pigeonpea (International Board for Plant Genetic Resources and International Crops Research Institute for the Semi-Arid Tropics, 1993). The quantitative traits were: days to 50% flowering, days to maturity, plant height (in centimeters), number of pods per plant, number of seeds per pod, number of seeds per plant, 100-gram weight (in grams), primary branches, secondary branches and grain yield

Variability of the qualitative traits was assessed by the calculation of frequencies of each modality in each character. The Shannon-Weaver diversity index (H') was computed using the phenotypic frequencies to assess the phenotypic diversity for each character for all accessions as described by Perry and McIntosh (1991). Each H' value was divided by its maximum value (logen) and normalized to keep the values between 0 and 1.

The diversity index (H') according to Shannon and Weaver (1949) is given in Equation 1:

$$H' = -\sum_{i=1}^{s} pi\ In\ (pi) \quad (1)$$

where: s is the number of phenotypic classes for a character and pi is the relative proportion of the total number of entries (N) in the i^{th} class.

Analysis of variance was done for the 16 quantitative traits over two sites using the Genstat 14th edition software (VSN International, 2011) after homogeneity of variances testing (Gomez and Gomez, 1984). The principal component analysis was also calculated using the Genstat 14th edition software and the bootstrap hierarchical cluster analysis was constructed based on Euclidean distances using the unweighted pair group method with arithmetic mean algorithm in the Genstat 20th edition software (VSN International, 2019).

Results

Variation in qualitative traits

Data for the frequency distribution for the 10 qualitative traits are presented in Table 2. Semi-spreading was the dominant growth

habit (97.9%) with only one erect/compact genotype and most pods were flat (97.9%), while one genotype had cylindrical pods (2.1%). There was variation in stem color with green (66.7), purple (17.0%) and dark green (17.0%) stems. Cream was the dominant seed color (89.6%), others had light grey (4.2%) and light brown (6.3%) seeds. The flowering patterns were semi-indeterminate (4.2%), determinate (89.6%) and indeterminate (8.3%). The highest variation was observed for flower color with yellow (39.6%), light yellow (31.3%), ivory (12.5%) green (10.42%) and red (6.25%). The seed coat patterns were plain (64.6%), speckled (27.1 %) and a few were mottled/speckled (8.33%). The seed color on the eye of the seed was highly variable from no color (83.3%), to light brown (4.17%), purple (4.2%) and reddish brown (4.2%).

The Shannon-Weaver diversity index (H') based on 10 qualitative traits revealed a low diversity among the genotypes studied (Table 2) ranging from 0.10 (pod form) to 1.04 (pod color). Pod color was the most diversified (1.04) followed by stem color (0.86) and the seed color pattern (0.84). In general, most of the studied qualitative traits were highly polymorphic. Some traits such as pod hairiness, leaflet shape, leaflet hairiness, pattern of streak and seed shape showed monomorphic characters and were excluded from the analysis.

Variation in quantitative traits

The results from the combined analysis of variance are presented in Table 3. There were significant ($p < 0.05$) differences between genotypes for the number of seeds per pod, 100-grain weight, number of branches, pod length, stem diameter, leaf length, leaf width and leaf area. There were significant ($p < 0.01$) differences between genotypes for the number of pods per plant, number of seed per plant and number of racemes. Highly significantly ($p < 0.001$) differences between genotypes were observed for the days to 50% flowering, days to maturity, plant height and grain yield. Data for mean, minimum and maximum values for the 16 quantitative traits are displayed in Table 4.

Table 2 Frequency and Shannon-Weaver diversity index of qualitative traits of 48 pigeonpea genotypes

Trait	Category	Frequency	Shannon-Weaver diversity index
Stem color	Green	32 (66.7%)	0.86
	Purple	8 (17%)	
	Dark green	8 (17%)	
Base flower color	Ivory	6 (12.5)	0.47
	Yellow	19 (39.6%)	
	Light yellow	15 (31.3%)	
	Green	5 (10.4%)	
	Red	3 (6.3%)	
Flowering pattern	Determinate	2 (4.2%)	0.13
	Semi-determinate	43 (89.6%)	
	Indeterminate	3 (6.3)	
Seed color pattern	Plain	31 (64.6%)	0.84
	Speckled	13 (27.1%)	
	Mottled/Speckled	4 (8.3%)	
	Mottled		
Seed eye color	None	40 (83.3%)	0.55
	Light brown	2 (4.2%)	
	Purple	2 (4.2%)	
	Reddish brown	2 (4.2%)	
	Grey/dark		
	Cream	2 (4.2%)	
Pod color	Purple	6 (12.5%)	1.04
	Green	30 (62.5%)	
	Black	3 (6.3%)	
	Mix	9 (18.8%)	
Pod form	Flat	47 (97.9%)	0.10
	Cylindrical	1 (2.1%)	
Base seed color	Cream	43 (89.6%)	0.13
	Dark purple		
	Light grey	2 (4.2%)	
	Light brown	3 (6.3%)	
Seed eye width	Narrow	20 (41.7%)	0.34
	Medium	28 (58.3%)	
	None		
	Wide		
Growth habit	Erect and compact	1 (2.1%)	0.31
	Semi-spreading	47 (97.9%)	

Table 3 Analysis of variance for 16 quantitative traits of pigeonpea

Trait		DT50F		DTM		PH		NPP	
Source	DF	MS	F-value	MS	F-value	MS	F-value	MS	F-value
Environment (E)	1	49,667.89	1884.93***	29,957.40	660.87***	607,484.91	827.77***	960,037.91	57.18***
Genotype (G)	47	494.89	18.78***	750.16***	16.55***	1,678.59	2.28***	32,347.47	1.93**
G×E	47	144.73	5.49***	130.88***	2.88***	1,571.39	2.14^{ns}	26,085.88*	1.55*
Error	71	26.35		45.33		733.88		16,790.64	
Mean		115.05		162.09		200.95		247.96	
CV		4.4		4.15		13.48		52.26	
Trait		NSOP		NSP		100-gw		NPB	
Source	DF	MS	F-value	MS	F-value	MS	F-value	MS	F-value
Environment (E)	1	4.86	6.39**	19599301.48	47.13**	133.37	32.93**	3275.66	4.87*
Genotype (G)	47	1.95	1.95*	822118.86	1.98**	28.36	7.01*	1275.62	1.89*
G×E	47	1.21	1.21**	580108.83	1.39^{ns}	16.29	4.02^{ns}	786.59	1.17^{ns}
Error	71	0.76		415836.5		4.05		672.94	
Mean		5.76		1397.21		19.68		38.78	
CV		13.27		46.15		20.57		72.31	
Trait		NSB		GY		PL		SD	
Source	DF	MS	F-value	MS	F-value	MS	F-value	MS	F-value
Environment (E)	1	80512.33	30.93***	10252919.86	58.28***	22.54	14.00***	529.71	246.38***
Genotype (G)	47	4491.38	1.72*	519644.12	2.95***	2.42	1.50*	4.07	1.89*
G×E	47	3420.15	1.31^{ns}	227358.82	1.29^{ns}	1.86	1.15^{ns}	2.26	1.05*
Error	71	2602.6		175918.66		1.61		2.15	
Mean		85.07		1237.65		7.8		8.4	
CV		59.97		33.89		17.4		24.01	
Trait		LFL		LW		LA		RCM	
Source	DF	MS	F-value	MS	F-value	MS	F-value	MS	F-value
Environment (E)	1	0.59	0.35^{ns}	0.03	0.08^{ns}	1085.17	5.16**	11395.31	52.83***
Genotype (G)	47	2.81	1.68*	0.62	1.55*	394.27	1.87*	415.69	1.93**
G×E	47	1.69	1.01^{ns}	0.45	1.13^{ns}	226.67	1.08^{ns}	216.68	1.01^{ns}
Error	71	1.67		0.4		210.47		215.71	
Mean		8.01		3.53		48		31.34	
CV		16.12		17.96		30.22		46.97	

*, **, *** = significant at $p < 0.05$, $p < 0.01$ and $p < 0.001$, ns-non-significant, respectively

DF = degrees of freedom, MS = mean square, F-value; DT50F = days to 50% flowering; DTM = days to maturity; NPP = number of pods/plant; NSOP = number of seeds/pod; NSP = number of seeds/plant; 100-gw = 100-grain weight; NPB = number of primary branches; NSB = number of secondary branches; GY = grain yield; PL = pod length; SD = stem diameter; LFL = leaflet length; LW = leaflet width; LA = leaf area; RCM = number of racemes; CV = coefficient of variation

Table 4 Descriptive statistics for 16 quantitative traits of pigeonpea

Trait	Mean	Minimum	Maximum	CV
Days to 50% flowering	115	81	149.5	12.5
Days to Maturity	162	126	200	10.51
Plant height (cm)	200	90	239	13.33
Number of pods/plant	246	103	561	39.56
Number of seeds/pod	576	3.95	6.95	10.78
Number of seeds/plant	1372	570.9	2435	34.51
100-gram weight (gm)	19.67	12.52	25.97	16.96
Number of primary branches	39.62	3.4	135.2	49.05
Number of secondary branches	86.84	3.03	231	45.62
Grain yield (kg)	1241	560.3	2030	32.38
Pod length (cm)	7.82	5.59	9.917	11.69
Stem diameter (cm)	8.38	6.168	11.17	12.47
Leaflet length (cm)	8.011	5.607	10.79	15.21
Leaf width (cm)	3.536	2.64	5.27	21.26
Leaf Area (cm^2)	48	22.75	81.82	30.44
Number of racemes	31.35	13.26	58.55	35.88

CV = coefficient of variation

Relationships among agro-morphological characters

The mean values from the two experimental sites were used to predict the relationship between the quantitative traits. Of the 16 quantitative traits, 6 high positive and significant correlations were observed (Table 5). The highest correlation coefficient (r = 0.99) was recorded between days to 50% flowering and days to maturity, followed by grain yield and number of seeds per plant (r = 0.93), number of primary branches and number of secondary branches (r = 0.85), leaf length and leaf width (r = 0.83), number of seeds per plant and number of seeds per pod (r = 0.75) and number of pods per plant and grain yield (r = 0.72). These values indicated that all yield components were important for grain yield. However, weak positive correlations were recorded between grain yield and stem diameter (r = 0.001) and grain yield and 100-grain weight (r = 0.01). Negative associations were observed between some pairs of characters. In total, 25 negative correlations were detected of which 18 pairs were not significant, 7 pairs were significant ($p < 0.05$) and the association between leaflet width and number of racemes was highly significant ($p < 0.01$).

Principal component analysis and cluster analysis

The principal component analysis (PCA) was based on 16 quantitative traits and their results are given in Table 6. The first four principal components with eigenvalues greater than 1.0 together accounted for about 73.4 % of the total variation among the collection (Table 6). The relative discriminating power of the principal axes as indicated by the eigenvalues was high (5.3%) for axis 1 and low (1.2%) for axis 4. The first principal component (PC1) explained 32.8% of the variation and was correlated mainly with days to 50% flowering, days to maturity, grain yield, leaf area and leaf width.

The second principal component (PC2) was responsible for about 23.8% of the variation and was negatively correlated with number of pods per plant, number of primary branches, number of secondary branches and grain yield. Moderate associations were observed between PC2 and the characters of pod length and leaf width. The proportion of variance explained by the third principal component (PC3) was 9.3% and was correlated with the number of primary branches and the number of secondary branches, indicating that PC3 was determined by vegetative parameters. The fourth principal component (PC4) accounted for 7.6% of the variation and was correlated with plant height and 100-grain weight.

Days to 50% flowering, days to maturity, number of seeds per pod, number of seeds per plant, grain yield, pod length, stem diameter, leaflet length, leaf width and leaf area were considered the most important for the characterization of pigeonpea germplasm, as they appeared in the four principal components three times. The remaining characters made no contribution to the variation in the four PCs and therefore were of minor importance in the characterization of pigeonpea germplasm.

The clustering carried out on the principal components grouped the 48 pigeonpea genotypes into three clusters (Fig. 2). The clustering pattern revealed that genetic diversity was not associated with geographical diversity. Cluster 1 contained mostly improved genotypes collected from Kenya and local landraces from Tanzania and was characterized by medium and late duration cycles with more branches. Cluster 2 consisted of genotypes characterized by early, medium and late duration cycles. Most genotypes in this cluster had less branching with short and intermediate plant statures and low and high seed weights. Cluster 3 contained mostly landrace genotypes and few improved genotypes. The individuals in this cluster could be characterized by medium and late maturing cycles, medium to high plant statures and high numbers of primary and secondary branches, number of seeds per plant, number of pods per plant and grain yield. In this study, the genotypes Bangili, TZA 5463, Babati White, ICEAP 00040, ICEAP 00932, ICEAP 00557 and ICEAP 00554 were selected because they had desirable traits that could be utilized in a future breeding program.

Discussion

The studied 48 pigeonpea genotypes showed significant variability in agro-morphological traits as evident from various statistical parameters. The most important qualitative traits responsible for variability observed in the current study were stem color, growth habit, flowering pattern, seed eye color, base flower color, pod from, base seed color and seed color pattern. Green was the dominant stem color followed by purple and dark green. The dominance of the green stem color reported in this study agreed with the results of Upadhaya et al. (2005) and Manyasa et al. (2008). However, in contrast to the current results, Saxena and Sharma (1990) in a similar study reported the dominance of purple stem color on African landraces.

Semi-determinate was the dominant growth habit (97.9%). The preponderance of the semi-determinate growth habit in Tanzania pigeonpea germplasm was also reported by Manyasa et al. (2008). Upadhaya et al. (2014) reported that 91% of a 1,290 pigeonpea collection from the Caribbean and Central American regions consisted of semi-determinate genotypes. This result was in contrast to that of Zavinon et al. (2019) who reported the dominance of compact and erect (55.68%) genotypes for their Beninese pigeonpea landraces collection. It has been reported that farmers in Africa do not prefer the semi-determinate types because they do not support the development of other crops cultivated in association. Semi-determinate was also the dominant flowering pattern. Several authors reported the dominance of an indeterminate flowering pattern in pigeonpea (Remanandan et al., 1988; Upadhaya et al., 2005; Manyasa et al., 2008; Upadhaya et al., 2014).

The pod color and form are important preferred traits to consumers and traders. According to Saxena et al. (2010), immature pods that have a good appearance fetch a good price in the market. The dominance of green color in pods observed in the current study supports the consumer preference for pigeonpea with green pods reported by several researchers (Saxena et al., 1983; Pandita and Dahiya, 1988; Upadhaya et al., 2010). Consumers of pigeonpea preferred green-colored pods because they believed this color remained attractive for 3–5 d after harvest.

The diversity was calculated using H' for all 10 qualitative traits. Based on this index, the pod form (0.10), flowering pattern (0.13) and base seed color (0.13) had low diversity, showing little variation among genotypes. This result was similar to that reported by Upadhaya et al. (2005). The H' value for pod color (1.04) was higher than that of 0.75 reported by Zavinon et al. (2019) and less than that of 1.39 reported by Ayenan (2017). H' for stem color (0.86) recorded in the current study was similar to the value of 0.87 reported by Zavinon et al. (2019) and this latter researcher also reported higher H' values for growth habit, seed color pattern and pod form for Beninese pigeonpea. Comparable results were produced by Ayenan (2017) with high diversity index values for 12 traits and low diversity for pod shape (0.39) and no variation for pod hairiness. The results observed in the current study showed that West Africa pigeonpea germplasm had a higher diversity index than East Africa germplasm, perhaps because the germplasm had undergone little selection, with low introduction of improved varieties showing more uniformity.

Table 5 Correlation coefficient for 16 quantitative traits of pigeonpea

	Trait	1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16
1	DT50F	-															
2	DTM	0.99***	-														
3	PH	0.52***	0.51***	-													
4	NPP	0.37*	0.39**	0.14	-												
5	NSOP	0.54***	0.51***	0.12	0.17	-											
6	NSP	0.48***	0.53***	0.25	0.75***	0.39*	-										
7	GW	-0.15	-0.12	0.16	0.06	-0.14	0.08*	-									
8	NPB	0.06	0.09	0.11	0.51***	-0.11	0.31*	0.14	-								
9	NSB	-0.12	-0.11	0.08	0.25	-0.12	0.17	0.15	0.85***	-							
10	GY	0.55***	0.58***	0.25	0.72***	0.35*	0.93***	0.01	0.32*	0.17	-						
11	PL	0.26*	0.22	0.18	0.11	0.17	-0.23**	-0.38*	-0.29*	-0.37*	-0.21	-					
12	SD	0.45*	0.40**	0.24	0.21	0.29*	0.13	-0.27	0.03	-0.11	0.19	0.46**	-				
13	LFL	0.50***	0.47***	0.12	0.04	0.26	0.02	-0.41*	-0.06	-0.21	0.00	0.52***	0.49***	-			
14	LW	0.60***	0.56***	0.24	0.07	0.30*	-0.01	-0.44*	-0.12	-0.30*	0.07	0.52***	0.57***	0.82***	-		
15	LA	0.54***	0.49***	0.17	0.33*	0.46***	0.25	-0.06	0.08	0.03	0.35	0.23	0.54***	0.31*	0.56***	-	
16	RCM	0.20	0.18	0.13	0.61***	0.15	0.51***	0.08	0.26	0.26	0.56***	-0.18	-0.03	-0.21	-0.13***	0.16	-
		1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16

*, **, *** = significant at $p < 0.05$, $p < 0.01$ and $p < 0.001$, respectively
Abbreviations are the same as shown in table 3.

Table 6 Principal component analysis for 16 quantitative traits

	PC1	PC2	PC3	PC4
Eigenvalue	5.245	3.72	1.484	1.217
Proportion of variance (%)	32.78	23.75	9.28	7.61
Total variance (%)	32.78	56.53	65.81	73.42
Eigenvector (loading)				
Days to 50% flowering	**0.396**	0.044	0.103	-0.217
Days to maturity	**0.388**	0.018	0.120	0.230
Plant height	0.197	-0.033	0.035	**-0.687**
Number of pods/plant	0.254	**-0.308**	-0.062	0.224
Number of seeds/pod	0.260	0.043	0.248	0.136
Number of seeds/plant	0.279	-0.313	0.204	0.119
100-grain weight	-0.099	-0.230	0.187	**-0.464**
Number of primary branches	0.075	**-0.332**	**-0.569**	-0.088
Number of secondary branches	-0.014	**-0.331**	**-0.546**	-0.107
Grain yield	**0.300**	**-0.300**	0.176	0.143
Pod length	0.136	**0.351**	-0.099	0.005
Stem diameter	0.261	0.178	-0.233	0.160
Leaflet length	0.247	0.294	-0.237	-0.058
Leaf width	**0.285**	**0.310**	-0.184	0.052
Leaf area	**0.297**	0.049	-0.104	0.088
Number of racemes	0.145	**-0.315**	0.116	0.226

PC = principal component

Values in bold made substantial contributions to total variation

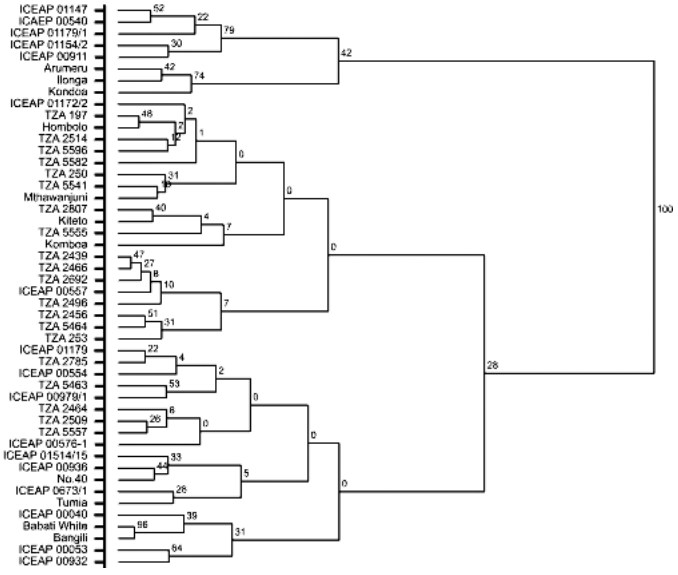

Fig. 2 Unweighted pair group method with arithmetic mean (UPGMA) dendrogram showing phenotypic cluster of 48 pigeonpea genotypes based on 16 quantitative traits, with bootstrap values (%) based on 1,000 permutations

The combined analysis of variance revealed significant variation for the different quantitative traits. According to Upadhaya et al. (2005), important quantitative traits to characterize pigeonpea germplasm include days to maturity, plant height, number of pods per plant, number of seeds per pod, number of racemes, number of secondary branches and pod length. The number of days to 50% flowering was in the range 81–149 and days to maturing was in the range 126–200 d, as shown in Table 4. The current results agreed with Zavinon et al. (2019) who recorded a range of 132.5–228 d with a mean of 192 d. The early duration genotypes such as Komboa (81 d) evaluated in the current study are best suited for the dry areas in the central and some western parts of Tanzania. This genotype is an excellent candidate material for the development of early maturing genotypes.

A high number of seeds per pod has been important in areas where pigeonpea is mainly grown for the domestic market and consumed as a green vegetable. This was in agreement with observations by Omanga et al. (1995) and Shiferaw et al. (2007), who reported preferences of farmers for a high number of seeds per pod and seed weight (100-grain weight). A similar observation was also reported by Saxena et al. (2010) regarding strong consumer preference for genotypes with many seeds per pod. The high number of seeds per pod observed in the current study confirmed the findings by Silim et al. (2006), Upadhaya et al. (2005) and Manyasa et al. (2008), who reported higher numbers of seeds per pod in the African germplasm than in the Indian germplasm.

Long duration genotypes had higher numbers of primary and secondary branches than medium and short duration genotypes because of their semi-spreading growth habit. According to Baldev (1988), the semi-spreading, long duration genotypes had a higher number of branches than the medium and early maturing genotypes. The high values observed in the numbers of primary and secondary branches could also be explained by the large inter-row and intra-row spacing adopted in the current study. Mula et al. (2011) reported that at a low population density, pigeonpea produced greater biomass. The current study suggested the genotypes with a short duration cycle to reach maturity and high number of seeds per pod could be crossbred with those with higher yield but with long maturity. The results may be particularly important for breeders and farmers to develop varieties with high yield potential.

The correlation coefficient between traits ranged from strong positive to strong negative. Strong positive correlation coefficients

were found between days to 50% flowering and days to maturity, number of primary and secondary branches, leaf length and leaf width, number of seeds per pod and number of seeds per plant, and number of pods per plant and grain yield. The findings from the current study were similar to those reported by Sreelakshmi et al. (2011). Upadhaya et al. (2014) reported high correlations between days to 50% flowering and days to 75% maturity and between number of seeds per plant and number of pods per plant. According to Singh et al. (1990), a strong correlation between yield and yield components would imply that it would be possible to improve both traits simultaneously. The 100-grain weight and number of primary branches had a weak positive correlation. This finding contrasted with what was observed by Musaana and Nahdy (1998), who reported a strong positive correlation.

A negative correlation was recorded between leaf width and 100-grain weight, while a weak negative correlation was observed between number of seeds per plant and leaf width, number of racemes and stem diameter, number of seeds per pod and number of primary branches. The negative correlation between number of seeds per pod and number of primary branches implied that the number of primary branches was not an important yield component. The same observation was reported by Musaana and Nahdy (1998). In the current study, the days to 50% flowering, days to maturity, number of seeds per pod, number of seeds per plant, number of pods per plant, grain yield, number of primary and secondary branches, leaf length and leaf width were identified as selection criteria for obtaining good parents in a pigeonpea breeding program.

From PCA, the most important traits to distinguish pigeonpea genotypes were days to 50% flowering, days to maturity, grain yield, leaf width, leaf area, number of seeds per plant, stem diameter, number of seeds per pod and number of pods per plant. Using PCA, some of these traits have also been used extensively to characterize pigeonpea (Upadhaya et al., 2007; Manyasa et al., 2008; Rao, 2009; Rekha et al., 2013, Zavinon et al., 2019).

The hierarchical cluster analysis conducted on the 16 quantitative traits grouped the genotypes into three clusters, indicating sufficient variability to warrant selection. Manyasa et al. (2008) classified 123 pigeonpea genotypes into six clusters. The grouping of pigeonpea into six clusters has been reported by Birhan et al. (2013) and Rupika et al. (2014) who observed 100 and 90 genotypes, respectively, grouped into six clusters. Shunyu et al. (2013) observed 30 pigeonpea genotypes grouped into 7 phenotypic clusters. Ayenan (2017) observed 49 pigeonpea genotypes grouped into 10 phenotypic clusters. In the current study, it was observed that clustering of genotypes from different countries into one cluster could be attributed to the exchange of breeding material. Furthermore, material may be selected from clusters for crosses that would result in high genetic gain. Thus, accessions from cluster III were the potential sources of genetic material for breeding varieties combining superior agronomic traits.

The study revealed the existence of considerable variation among the germplasm accessions. The PCA identified days to 50% flowering, days to maturity, number of pods per plant, number of seed per plant, grain yield, leaf width and leaf area as the most important traits for characterization in pigeonpea improvement. Furthermore, the cluster analysis grouped the genotypes into three clusters. The genotypes Bangili, TZA 5463, Babati White, ICEAP 00040, ICEAP 00932, ICEAP 00557 and ICEAP 00554 had desirable phenotypic attributes and should be utilized in further breeding programs for developing superior varieties.

Conflict of Interest

The authors declare that there are no conflicts of interest.

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