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Agro-morphological characterization of pigeonpea (*Cajanus cajan* L. Millspaugh) landraces grown in Benin: Implications for breeding and conservation

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Pigeonpea (*Cajanus cajan* L. Millspaugh) is a neglected and under-utilized crop consumed in several regions of word. In order to assess performance of pigeonpea landraces grown in Benin for useful breeding programs, 50 accessions were collected from 39 villages. These accessions were characterized by using 12 qualitative and 11 quantitative traits. Based on the seeds morphological characteristics, the 50 accessions were grouped in 12 morphotypes. However, 8 morphological classes were obtained with cluster analysis based on the unweighted pair group method with arithmetic average method using qualitative traits, whereas in principal component analysis only 5 clusters have been obtained using quantitative traits. The association/correlation among quantitative characters showed that grain yield was negatively correlated with pod width, days to 50% flowering and physiological maturity while it was positively correlated with pod length, pods per plant, branches per plant and number of seeds per pod. Based on four quantitative traits (number of pods per plant, number of seeds per pod, 100 seed weight, and early maturity), the 23 accessions from cluster 3 of whom kk5 (*Ekloui*), kk8 (*Nontchiovi kloui*), kk15 (*Otili founfoun*), kk18 (*Klouékoun wéwé*), kk22 (*Otili*), kk23 (*CA monlikoun*) and kk28 (*Hounkoun wéwé*) have been recommended as good sources of germplasm for improving the pigeonpea productivity. Further characterization using molecular techniques as well as conservation attention should be conducted to confirm the present result and maintain the germplasm for future breeding programs.

Keywords: Benin, Cluster analysis, morphological diversity, pigeonpea, quantitative characters, selection.

INTRODUCTION

Pigeonpea (*Cajanus cajan* L. Millspaugh) is a shrub, which plays an important role in food security, nutritional balance and poverty alleviation in sub-Saharan Africa (Rao et al., 2002). It is predominantly cultivated in the developing countries of tropical and subtropical environments (Suman et al., 2017). Africa, with 19.03% of the world's total production represents the second producer followed by Americas (3.15%) and behind Asia (77.82%) (Anon, 2017a). In Benin, though this legume is not considered by farmers as a priority crop, pigeonpea is the sixth-largest legume crop with a cultivated area of 3027 ha with an average yield of 1843 tons, behind groundnut, cowpea, soybeans, bambara groundnut and Kersting's groundnut (Anon, 2017b).

Various parts of pigeonpea plant are used for food consumption, as medicine for cure diseases. Leaves are used in traditional medicine to cure diseases such as malaria and fever, in Benin (Dansi et al., 2012; Ayenan et al., 2017; Zavinon et al., 2018), in Nigeria (Aiyéloja and Bello, 2006; Oladunmoye et al., 2011) and in South Africa (Mander et al., 1996). In most African countries, seeds are used in human nutrition as food in combination with cereals and in commercialization (Odeny, 2007; Dansi et al., 2012; Ayenan et al., 2017). In Benin, seeds are highly consumed in the Adja cultural area in the South-East (Dansi et al., 2012). Pigeonpea also has a strong potential to contribute to food security through market possibilities and by using it to make up for the shortage of cowpea, maize and other staple foods during lean season (Ayenan et al., 2017). The plant is also useful in soil conservation and weed management (Versteeg and Koudokpon, 1993; Aihou, 2003; Dansi et al., 2012).

The potential yield of pigeonpea is estimated at 2500 kg/ha, while the yields obtained on farmer's fields is estimated at 736.2 kg/ha in Africa and 620 kg/ha in Benin (Dutta et al., 2011; Anon, 2017b). The relatively lower yield obtained is due to biotic and abiotic constraints and as well lack of quality seed (Ayenan et al., 2017). Moreover, these constraints can cause yield penalty of pigeonpea and could be involved in the long term process disappearance of some landraces. In fact, the evaluation of genetic diversity is essential for efficient use and conservation of pigeonpea genetic resources (Shende and Raut, 2013). It is therefore important to know genetic variability among pigeonpea landrace in Benin for future breeding research and conservative management.

In Benin, various landraces of pigeonpea are grown across different ecological zones and their vernacular names were given by farmers to distinguish them. However, pigeonpea's vernacular names usually vary from one ethnic group to another, from one village to another within the same ethnic area and sometimes from one household to another within the same village (Ayenan et al., 2017). In this context a cultivar across villages may be designated by different names while different cultivars can sometimes be designated by the same name (Otoo et al., 2009; Agre et al., 2015). For instance, in the Guinean and Sudano-Guinean zones of Benin, pigeonpea is called Hounkoun, Kloué or Klouékoun by farmers belonging to Fon and Mahi sociolinguistic groups while in the Guinean and Sudanian zones, pigeonpea is called Otili by farmers belonging to Nago and Dendi sociolinguistic groups (Kinhoégbè et al., 2019). This constitutes a bias in the estimation of pigeonpea diversity. Characterization of existing landraces germplasm is a prerequisite step for identifying potential germplasm to be used in breeding program and also avoid duplication in the germplasm collection.

Different methods can be used to access genetic variability in plant species, such as pedigree data, morphological and molecular markers. The use of agro-morphological traits is the most common approach utilized to estimate relationships between genotypes and provide information for plant breeding programs (Bajracharya et al., 2006; De, 2019). Data obtained by landrace description are futher statistically processed. Multivariate analysis such as cluster analysis, Principal Component Analysis (PCA) and discriminate analysis is the most commonly used approach for genetic variability estimation to illuminate the patterns of variation in germplasm collections. Among multivariate techniques, PCA and cluster analysis are preferred tools for morphological characterization of genotypes and their grouping on similarity basis (Mohammadi and Prasanna, 2003). Cluster analysis is used to reveal the association between landraces while relationships between traits are statistically analyzed using PCA. Landraces can be grouped together based on informative data and be used directly in a breeding program. In Africa, many studies have been conducted to examine patterns of genetic diversity among pigeonpea accessions using both qualitative and quantitative agro-morphological descriptors (Silim et al., 2005; Manyasa et al., 2008; 2009; Gwata and Slim, 2009; Vange and Egbe, 2009; Kundy et al., 2015). Unfortunately, in Benin, very scarce study has been done to characterize pigeonpea landraces (Quenum et al., 2016). This study however based on the evaluation of the pigeonpea seeds quality, allowed a partial characterization of the plants of the different morphotypes consequently, different landraces agronomic performances were not evaluated and conservation strategy of this genetic resource has not been developed in Benin. The objectives of this study were to classify the different pigeonpea landraces under cultivation in Southern and Central region of Benin and evaluate the agronomic performance of these accessions.

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MATERIALS AND METHODS

Description of experimental site

The present study was carried out in the experimental site of the Laboratory BIORAVE (Center for Research, Training, Incubation, Technological Innovation and Seed Production for Agricultural Development) at Massi (9°55'0" N and 1°28'0" E) in the municipality of Zogbodomè (Benin Republic) during the cropping season of 2017 to 2018 (April 2017 to January 2018). The site benefits a sub-equatorial climate with two dry seasons and two rainy seasons. The long rainy season extends from March to July and the short one from September to November. As for the dry seasons, they cover the period from December to March, and from July to September (Adam and Boko, 1993). The average annual temperature varies between 26 and 28°C (Yabi and Afouda, 2012) and the annual rainfall varies between 800 and 1,200 mm (Adam and Boko, 1993). The soil is ferruginous type dominated by sandy-clay sediments.

Plant material

The study was carried out on 50 accessions of pigeonpea, collected from 39 villages belonging to 7 different ethnic groups located in the departments of Southern and Central part of Benin (Kinhoégbè et al., 2019). In fact, 54 accessions were collected during an ethnobotanical survey and according to famers seem to have different agronomical performances. From these 54 accessions, four did not germinate and data were collected on 50 accessions that germinated during the experiment. Among these accessions, 29 were collected from Central region and 21 from South (Table 1).

Field layout

The experimental design used was randomized complete block (RCBD) with three repetitions. We used tree blocks of 50 plots corresponding to the 50 pigeonpea accessions. Plots were 11 m length with 1.5 m and 1 m row spacing. At the time of sowing, three seeds were put in a pouch. The depth of sowing was 3cm. After 30 days, extra plants were removed and the most healthier and vigorous plants were left for phenotyping. The experiment was carried out without application of fertilizer since the soil is naturally fertile enough to support the crop.

Morphological traits/characters studied

Firstly, seed classification was made based on seed's morphological description characteristics (seed colour pattern, seed colour, seed eye colour, seed shape and seed size as described in Loko et al. (2018). Secondly, a total of 23 characters including 12 qualitative (Table 2), 11 quantitative (Table 3) were recorded according to the descriptors of *C. cajan* recommended by IBPGR and ICRISAT (1993). The different traits: plant height (PlHe), stem thickness (StT), branches per plant (BrP), pod length (PL), pod width (PWi), number of pods per plant (PPl), number of seeds per pod (SP), grain yield (GY), 100-seed weight (100SW), days to 50% flowering (D50F), physiological maturity (PhM), growth habit (GH), leaflet shape (LSh), base flower colour (BFCo), pod colour (PCo), pod colour pattern PCoPa), pod shape (PSh), pod form (PFo), seed shape (SSh), seed colour pattern (SCoPa), seed colour (SCo), seed eye colour (SECo), and seed size (SSi); were measured from vegetative stage until harvest according to the nature of each trait. For instance, growth habit and leaflet shape were recorded at preflowering while the base flower colour was recorded at flowering. Seed colour pattern, seed size and seed colour were recorded at the harvest of dried seeds, plant height, stem thickness and branches per plant at the end of flowering, number of pods per plant and number of seeds per pod at the first and second harvest of dried seeds (Tables 2 and 3). Data were recorded on five plants randomly selected from the eight planted in each row except the bordering plants in each row.

Data analysis

To group accessions with homogeneous morphological class, the genetic distance between accessions was calculated according to Nei (1972). The distance matrix obtained served for the construction of a dendrogram by the UPGMA (Unweighted Pair Group Method with Arithmetic average) method using SAHN (Sequential Agglomerative Hierarchical Nested) clustering of the NTSYS-pc software (Rohlf, 2000). Subsequently, using Minitab 16 software, the quantitative characters were initially subjected to a descriptive statistic and secondly to see relation between pairs of quantitative characters, Pearson correlation coefficient was performed. To examine the contribution of each quantitative character to total genetic variation, Principal Component Analysis (PCA) was performed. Then, on the basis of the Principal Component Analysis (PCA), accessions were projected on the first two PCs, in order to group different accession into clusters. In order to determine the differences in performance of the landraces for each agronomic trait, analysis of variance (ANOVA) was performed by using Minitab 16 software. Significant differences between means were observed using Turkey test ($p < 0.05$) (Sangseok and Dong, 2018).

RESULTS

Distribution of phenotypic characters

The 50 accessions were classified in twelve (12) morphotypes according to the seed morphological description characteristics (Figure 1). The number of accession for each group, the accessions and their characteristics are presented in Table 4. Based on this classification, the majority of pigeonpea cultivar grown were of cream seed colour. The analysis of the variability of qualitative characters showed that all the evaluated characters were polymorphic (Table 5). From the results, 34 accessions showed semi-spreading growth habit and 48 lanceolated leaflet shape. Thirty-six landraces showed light yellow colour for base flower and 34 had green pod colour. Sixteen landrace showed right pod shape, 10 cylindrical pod form and 43 oval seed shape. Forty-two showed plain seed colour pattern and 42 accessions showed cream seed colour. Thirty and thirty-nine accessions showed red eye colour and intermediate size, respectively.

The characterization based on the 12 qualitative characters grouped the 50 accessions in 11 morphological type assembled in eight morphological classes named C1 to C8 (Figure 2).

- C1 (4 accessions) is characterized by erect growth habit, lanceolated leaflet, curved and flatted pod totally coloured in green containing oval and cream seeds.
- C2 (2 accessions) is characterized by erect growth

Table 1. List of the 50 studied pigeonpea accessions, their code, corresponding prospected village, districts, locality and sociolinguistic group where accession was collected.

N°	Local name	Codes	Villages	Districts	Localities	Sociolinguistic group
1	Adja Kloui	kk34	Fangnonhoué	Lalo	Southern	Adja
2	CA Monlikoun	kk23	N'gbèhouédo	Ouèssè	Central	Mahi
3	Carder Ekloui	kk33	Toimey	Klouékanmè	Southern	Adja
4	Carder Ekloui	kk35	Toimey	Klouékanmè	Southern	Adja
5	Ekloui	kk2	Dékpo	Aplahoué	Southern	Adja
6	Ekloui	kk5	Hélétoumey	Aplahoué	Southern	Adja
7	Ekloui	kk6	Hélétoumey	Aplahoué	Southern	Adja
8	Ekloui	kk38	Djowé	Aplahoué	Southern	Adja
9	Ekloui Ri	kk12	Golouhoué	Klouékanmè	Southern	Adja
10	Ekloui Ri	kk39	Golouhoué	Klouékanmè	Southern	Adja
11	Hounkoun Wéwé	kk28	Adaklamè-Dénou	Kétou	Southern	Mahi
12	Kloué	kk16	N'gbèhouédo	Ouèssè	Central	Mahi
13	Kloué	kk29	Atomey-Kpodji	Aplahoué	Southern	Adja
14	Kloué	kk36	Towé	Pobè	Southern	Yorouba
15	Klouékoun Vôvô	kk47	Hèlontèdji	Zangnannado	Central	Fon
16	Klouékoun Wéwé	kk4	Kpakpassa	Savalou	Central	Mahi
17	Klouékoun Wéwé	kk14	Soclogbo	Dassa-Zoumè	Central	Mahi
18	Klouékoun Wéwé	kk18	Kpakpassa	Savalou	Central	Mahi
19	Klouékoun Wéwé	kk20	Katakou	Savè	Central	Fon
20	Klouékoun Wéwé	kk37	Kèmondji	Zakpota	Central	Fon
21	Klouékoun Wéwé	kk40	Gossoé	Zangnannado	Central	Fon
22	Klouékoun Wéwé	kk41	Kèmondji	Zakpota	Central	Fon
23	Klouékoun Wéwé	kk43	Sohoungo	Zakpota	Central	Fon
24	Klouékoun Wéwé	kk44	Hounsso	Covè	Central	Fon
25	Klouékoun Wéwé	kk45	Hèlontèdji	Zangnannado	Central	Fon
26	Klouékoun Wéwé	kk46	Abahogo	Zangnannado	Central	Fon
27	Klouékoun Wéwé	kk48	Gossoé	Zangnannado	Central	Fon
28	Klouékoun Wéwé	kk49	Gossoé	Zangnannado	Central	Fon
29	Klouékoun Wéwé	kk50	Gbihoungon	Djidja	Central	Fon
30	Klouékoun wlanwlan	kk17	Monsourou	Djidja	Central	Fon
31	Klouékoun wlanwlan	kk27	Gbihoungon	Djidja	Central	Fon
32	Klouékoun wlanwlan	kk42	Gossoé	Zangnannado	Central	Fon
33	Nontchiovi Kloui	kk8	Hélétoumey	Aplahoué	Southern	Adja
34	Otili	kk22	Olata	Ouèssè	Central	Nago
35	Otili Founfoun	kk15	Towé	Pobè	Southern	Yorouba
36	Otili Founfoun	kk19	Oké-Ola	Kétou	Southern	Holly
37	Otili Founfoun	kk21	Chaffou	Pobè	Southern	Yorouba
38	Otili Founfoun	kk25	Ayétédjou	Dassa-Zoumè	Central	Holly
39	Otili Founfoun	kk32	Kèmon	Ouèssè	Central	Nago
40	Otili Founfoun Kékélé	kk7	Ferme Gbagba	Savè	Central	Biali
41	Otili Founfoun Lakoun	kk11	Monsourou	Djidja	Central	Fon
42	Otili Kpoukpa	kk13	Oké-Odja	Pobè	Southern	Yorouba
43	Otili Kpoukpa	kk30	Towé	Pobè	Southern	Yorouba
44	Otili Kpoukpa	kk31	Ayétédjou	Dassa-Zoumè	Central	Holly
45	Otini Founfoun	kk26	Ayétédjou	Dassa-Zoumè	Central	Holly
46	Otini Kpoukpa	kk1	Ayétédjou	Dassa-Zoumè	Central	Holly
47	Otini Kpoukpa	kk3	Oké-Ola	Kétou	Southern	Holly
48	Otini Kpoukpa	kk24	Oké-Ola	Kétou	Southern	Holly
49	Otini Tchofiti	kk9	Ayétédjou	Dassa-Zoumè	Central	Holly
50	Wlétchivé Kloui	kk10	Djowé	Aplahoué	Southern	Adja

Table 2. Qualitative morphological characters evaluated.

Character	Codes	Period of observation	Variables and score
Growth habit	GH	Preflowering	Erect (1) Semi-spreading (2) Spreading (3)
Leaflet shape	LSh	Preflowering	Oblong-lanceolate (1) Lanceolate (2)
Base flower colour	BFCo	Flowering	Light yellow (1) Yellow (2) Orange-yellow (3)
Pod colour	PCo	Harvest of green seeds	Green (1) Purple (2) Mixed (3)
Pod colour pattern	PCoPa	Harvest of green seeds	Total (1) Spots or bands dark rose (2) Pigmentation on the surface or in the cavities of the pod (3)
Pod shape	PSh	Harvest of green seeds	Right (1) Curve (2)
Pod form	PFo	Harvest of dried seeds	Flat (1) Cylindrical (2)
Seed shape	SSh	Harvest of dried seeds	Oval (1) Globular (2) Square (3)
Seed colour pattern	SCoPa	Harvest of dried seeds	Plain (1) Mottled (2)
Seed colour	SCo	Harvest of dried seeds	Cream (1) Blackish (2) Red (3) Brown (4) Light-red (5)
Seed eye colour	SECo	Harvest of dried seeds	Red (1) Black (2) No one (3)
Seed size	SSi	Harvest of dried seeds	Small (1) Intermediate (2) High (3)

habit, lanceolated leaflet, curved and flatted pod totally coloured in green containing globular and high cream seeds having red eyes.

- C3 (23 accessions) is characterized by erect growth habit, lanceolated leaflet, curved and flatted pod totally coloured in green containing globular and cream seeds having red eyes and intermediate size.
- C4 (5 accessions) is similar to the previous (C3) with the only difference by grouping seeds with small size.
- C5 (6 accession) is characterized by semi-spreading growth habit, lanceolated leaflet, light yellow base flower, right and flat pods having mixed colour with pigmentation on the surface or in their cavities, containing oval and mottled seeds having intermediate size.
- C6 (2 accessions) is characterized by spreading growth habit, oblong lanceolated leaflet, light yellow base flower, cylindrical and right pod having mixed colour with pigmentation on the surface or in theirs cavities, containing globular and mottled seeds having intermediate size
- C7 (3 accession) is characterized by semi-spreading growth habit, lanceolated leaflet, light yellow base flower, right and cylindrical pod shapes having purple colour with spots or bands dark rose, containing squared seeds entirely coloured in light-red having intermediate size.
- C8 (5 accessions) is characterized by semi-spreading growth habit, lanceolated leaflet, light yellow base flower, right and cylindrical pod having purple colour with spots

Table 3. Quantitative morphological characters evaluated.

Character	Code	Period of observation	Unit
Plant height	PlHe	End of flowering	m
Stem thickness	StT	End of flowering	mm
Branches per plant	BrPl	End of flowering	unity
Pod length	PL	Harvest of dried seeds	mm
Pod width	PWi	Harvest of dried seeds	mm
Days of 50% flowering	D50F	Flowering	Days
Physiological maturity	PhM	Physiological maturity	Days
Number of pods per plant	PPl	1st and 2nd harvest of dried seeds	unity
Number of seeds per pod	SP	1st and 2nd harvest of dried seeds	unity
Grain yield	GY	Harvest of dried seeds	tonnes/ha
100-seed weight	100SW	1st and 2nd harvest of dried seeds	g


G1 G2 G3
G4 G5 G6
G7 G8 G9
G10 G11 G12

Figure 1. Pictures of different groups obtained from seeds classification.

Table 4. Accessions corresponding of each group obtained based on morphological characteristics.

Group	Number of cultivars	Cultivar	Seed colour pattern	Seed colour	Seed eye colour	Seed shape	Size
G1	2	kk44; kk48	Plain	Cream	Black	Oval	Intermediate
G2	23	kk5; kk8; kk12; kk15; kk18; kk20; kk21; kk22; kk23; kk25; kk28; kk29; kk33; kk34; kk38; kk39; kk40; kk41; kk43; kk45; kk46; kk49; kk50	Plain	Cream	Red	Oval	Intermediate
G3	2	kk35; kk36	Plain	Cream	No one	Oval	High
G4	5	kk9; kk10; kk17; kk27; kk42	Highly mottled	Cream	No one	Oval	Intermediate
G5	1	kk11	Mottled	Cream	No one	Oval	Intermediate
G6	1	kk31	Plain	Brown	No one	Oval	Intermediate
G7	3	kk1; kk24; kk30	Plain	Red	No one	Oval	Intermediate
G8	3	kk3; kk13; kk47	Plain	Light red	No one	Square	Intermediate
G9	1	kk32	Plain	Blackish	No one	Oval	Intermediate
G10	5	kk4; kk7; kk14; kk16; kk37	Plain	Cream	Red	Oval	Small
G11	2	kk19; kk26	Plain	Cream	Red	Globular	High
G12	2	kk2; kk6	Mottled	Cream	No one	Globular	High

Table 5. Frequency of appearance of qualitative variables in set of collection

Character	Variables and score	Number of accession
Growth habit	Erect	14
	Semi-spreading	34
	Spreading	2
Leaflet shape	Oblong-lanceolate	2
	Lanceolate	48
Base flower colour	Light yellow	36
	Yellow	8
	Orange-yellow	6
Pod colour	Green	34
	Purple	8
	Mixed	8
Pod colour pattern	Total	34
	Spots or bands dark rose	8
	Pigmentation on the surface or in the cavities of the pod	8
Pod shape	Right	16
	Curve	34
Pod form	Flat	40
	Cylindrical	10
Seed shape	Oval	43
	Globular	4
	Square	3
Seed colour pattern	Plain	42
	Mottled	8
Seed colour	Cream	42
	Blackish	1
	Red	3
	Brown	1

Table 5. Contd.

	Light-red	**3**
Seed eye colour	Red	30
	Black	2
	No one	18
Seed size	Small	5
	Intermediate	39
	High	6

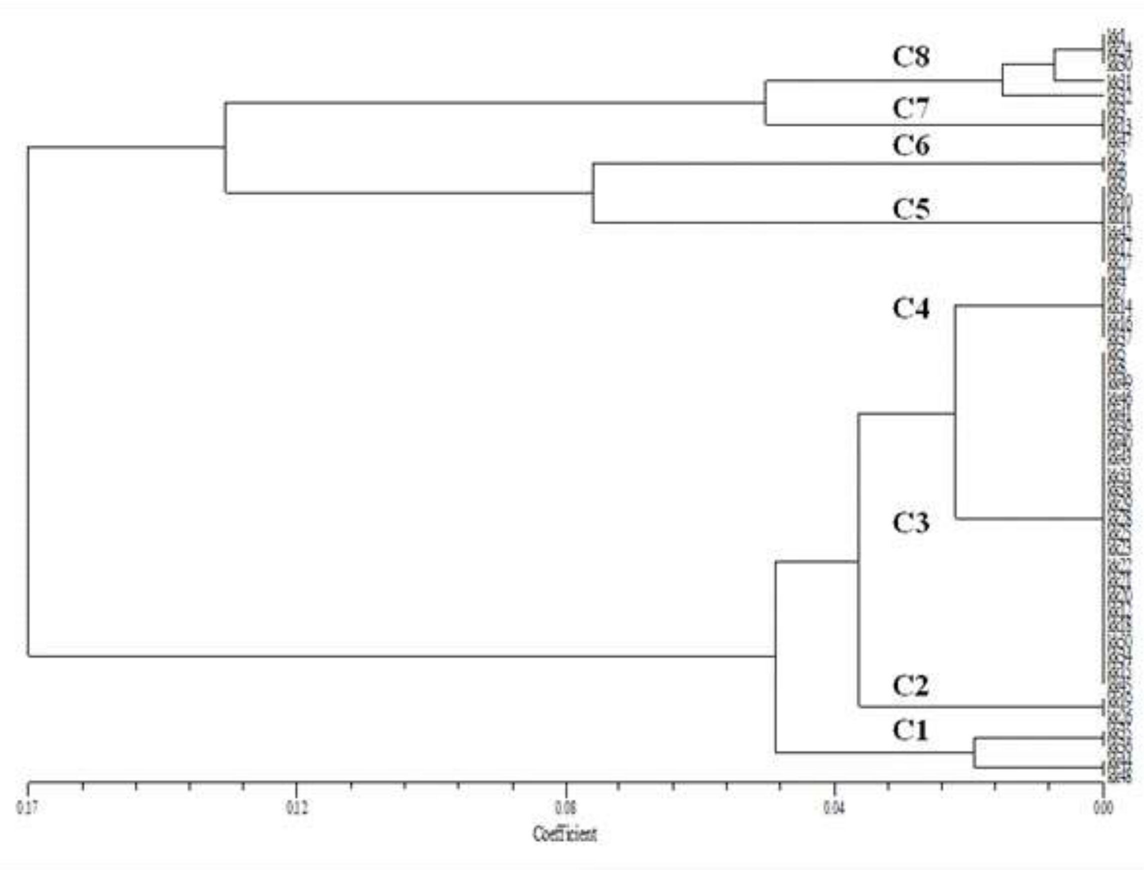

Figure 2. Dendrogram showing different morphological types assembly in morphological classes of pigeonpea in Benin using UPGMA method.

or bands dark rose, containing oval seeds entirely coloured, having intermediate size and without pigmentation and seeds eyes.

Agro-morphological evaluation based on quantitative traits

The results (Table 6) showed that branches per plant, number of pods per plant, pod width and grain yield were the most variable when referring to their coefficient of variation. The plant height ranged from 1.86 m (kk31) to 3.35 m (kk15) with an average of 2.93 m. The stem thickness ranged from 26.20 mm (kk31) to 66.20 mm (kk21) with an average of 51.93 mm. Mean number of branches per plant was 33.79 unities. The length of the pods ranged from 41.80 mm (kk17) to 71.33 mm (kk15), with an average of 61.74 mm and coefficient of variation of 16.06% while the width of the pods ranged from 3.48 mm (kk15) to 8.14 mm (kk19; kk26), with an average of 5.70 mm and a coefficient of variation of 37%. The number of pods per plant ranged from 134.60 unities

Table 6. Descriptive statistics of the quantitative characters evaluated.

Character	Mean	Min	Max	CoeffVar	StDev
PlHe	2.93±0.08	1.86	3.35	18.64	0.55
StT	51.93±1.93	26.20	66.20	26.32	13.67
BrPl	33.79±1.80	5.00	45.83	37.68	12.73
PL	61.74±1.40	41.80	71.33	16.06	9.92
PWi	5.70±0.30	3.48	8.14	37.00	2.11
PPl	1340.30±88.40	134.60	1956.30	46.62	624.80
SP	5.14±0.12	3.40	5.83	15.87	0.82
GY	3.73±0.17	0.55	4.74	32.55	1.21
100SW	10.84±0.21	7.54	12.50	13.84	1.50
D50F	135.21±3.95	109.00	185.00	20.67	27.94
PhM	174.77±2.78	156.00	228.00	11.24	19.64

Min: Minimal; Max: Maximal; CoeffVar: Coefficient of Variation; StDev: standard deviation; PlHe: Plant height; StT: Stem thickness; BrPl: Branches per plant; PL: Pod length; PWi: Pod width; PPl: Number of pods per plant; SP: Number of seeds per pod; GY: Grain yield; 100SW: 100-seed weight; D50F: Days of 50% flowering; PhM: Physiological maturity

Table 7. Correlation matrix among quantitative characters.

Character	PlHe	StT	BrP	PL	PWi	PPl	SP	GY	100SW	D50F	PhM
PlHe	1										
StT	0.96^{***}	1									
BrP	0.37^{*}	0.22^{ns}	1								
PL	0.92^{***}	0.82^{***}	0.58^{***}	1							
PWi	-0.54^{***}	-0.39^{*}	-0.89^{***}	-0.79^{***}	1						
PPl	0.91^{***}	0.94^{***}	0.42^{**}	0.82^{***}	-0.56^{***}	1					
SP	0.89^{***}	0.77^{***}	0.56^{***}	0.99^{***}	-0.79^{***}	0.76^{***}	1				
GY	0.98^{***}	0.95^{***}	0.47^{**}	0.89^{***}	-0.56^{***}	0.93^{***}	0.85^{***}				
100SW	0.14^{ns}	0.36^{*}	-0.43^{**}	-0.18^{ns}	0.48^{***}	0.34^{*}	-0.26^{ns}	0.19^{ns}	1		
D50F	-0.27^{ns}	-0.27^{ns}	-0.75^{***}	-0.34^{*}	0.67^{***}	-0.58^{***}	-0.28^{ns}	-0.39^{*}	-0.10^{ns}	1	
PhM	-0.46^{***}	-0.45^{**}	-0.77^{***}	-0.47^{***}	0.66^{***}	-0.70^{***}	-0.40^{**}	-0.58^{***}	-0.16^{ns}	0.95^{***}	1

PlHe: Plant height; StT: Stem thickness; BrP: Branches per plant; PL: Pod length; PWi: Pod width; PPl: Number of pods per plant; SP: Number of seeds per pod; GY: Grain yield; 100SW: 100-seed weight; D50F: Days to 50% flowering ; PhM: Physiological maturity; Significant correlations at $^{*}p < 0.05$, $^{**}p < 0.01$, $^{***}p < 0.001$; ns: not significant.

(kk32) to 1956.25 unities (kk15) with an average of 1340 unities. The mean of number of seed per pod was 5.14 unities. Grain yield ranged from 0.55 tons/ha (kk32) to 4.74 tons/ha (kk15; kk22 and kk25) with an average of 3.73 tons/ha. The 100-seed weight ranged from 7.54 g (kk4) to 12.5 g (kk19; kk21 and kk24) with an average of 10.84 g. The days to 50% flowering ranged from 109 days (kk15 and kk22) to 185 days (kk32) with an average of 135.21 days. Physiological maturity ranged from 156 days (kk20 and kk25) to 228 days (kk32) with an average of 174.77 days.

Correlation between/among quantitative characters

The coefficient of correlation between quantitative characters is presented in Table 7. The results showed that number of branches per plant (BrP) was positively correlated with pod length (PL) (r = 0.58^{***}), number of pods per plant (PPL) (r = 0.42^{**}), number of seeds per pod (SP) (r = 0.56^{***}) and grain yield (GY) (r = 0.47^{**}) while negatively correlated with pod width (PWi) (r = -0.89^{***}), 100-seed weight (100SW) (r = - 0.43^{**}), days to 50% flowering (r = -0.75^{***}) and physiological maturity (PhM) (r = -0.77^{***}). Pod length (PL) was positively correlated with number of seeds per pod (SP) (r = 0.99^{***}), number of pods per plant (PPL) (r = 0.82^{***}) and grain yield (GY) (r = 0.89^{***}) while it was negatively correlated with pod width (PWi) (r = - 0.79^{***}), days to 50% flowering (r = -0.34^{*}) and physiological maturity (PhM) (r = -0.47^{***}). Pod width (PWi) was negatively correlated with the number of pods per plant (PPL) (r = -0.56^{***}), the number of seeds per pod (SP) (r = -0.79^{***}) and grain yield (GY) (r = -0.56^{***}) while it is positively correlated with 100-seed weight (100SW) (r = 0.48^{***}), days to 50% flowering (r = 0.67^{***}) and physiological

Table 8. Correlations between characters and the first three factorial axes

Character	PC1	PC2	PC3
PlHe	0.34***	0.25**	0.14
StT	0.32***	0.36***	0.01
BrPl	0.26**	-0.46***	-0.05
PL	0.35***	0.04	0.27**
PWi	-0.31***	0.37***	-0.12
PPl	0.35***	0.20**	-0.15
SP	0.34***	0.02	0.34***
GY	0.35***	0.21***	0.03
100SW	0.00	0.48***	-0.54***
D50F	-0.23**	0.31***	0.50***
PhM	-0.28**	0.21**	0.46***
Eigen value	6.98	2.18	1.59
Proportion (%)	0.64	0.20	0.15
Cumulative proportion (%)	0.64	0.83	0.98

PlHe: Plant height; StT: Stem thickness; BrPl: Branches per plant; PL: Pod length; PWi: Pod width; PPl: Number of pods per plant; SP: Number of seeds per pod; GY: Grain yield; 100SW: 100-seed weight; D50F: Days of 50% flowering; PhM: Physiological maturity; *degree of correlative value with the axe.

maturity (PhM) (r = 0.66***). The number of pods per plant (PPL) was positively correlated with grain yield (GY) (r = 0.93***) while it was negatively correlated with days to 50% flowering (r = -0.58***) and physiological maturity (PhM) (r = -0.70***) while the number of seeds per pod (SP) was positively correlated with grain yield (GY) (r = 0.85***) and negatively with physiological maturity (PhM) (r = -0.40*). Grain yield (GY) was negatively correlated with physiological maturity (PhM) (r = -0.58***). Days to 50% flowering (D50F) was positively correlated with physiological maturity (PhM) (r = 0.95***).

Principal component analysis

The Principal Component Analysis performed using the 11 quantitative characters showed that the first two PC had an Eigen value higher than 1 and accounted for 83% of the total variability (Table 8). Plant height (PlHe), stem thickness (StT), branches per plant (BrPl), pod length (PL), number of pods per plant (PPl), number of seeds per pod (SP) and grain yield (GY) were positively correlated with PC1. The 100-seed weight (100SW) was negatively correlated with the 3rd PC and positively correlated with the 2nd axis. The correlation of the characters with the first two PCs is represented in Figure 3. The fifty accessions have been grouped in 5 clusters (Figure 4).

The landrace accessions of the cluster I (12 accessions; 3 from Central and 9 from Southern) are characterized by the high 100-seed weight (100SW) and pod width (PWi). The cluster III (23 accessions; 12 from Central and 11 from Southern) seems to group accessions with high good parameters of yield: pods per plant (PPl), number of seeds per pod (SP) and grain yield (GY). The cluster IV (5 accessions; all from Central) seems to group accessions witch maturing late. The cluster V (2 accessions, all from central) grouped accessions that have opposite performances to accessions of the cluster III. The cluster II (8 accessions; 7 from Central and 1 from Southern) group accessions with performance values close to the mean of those of fourth and cluster V.

The comparison of the means of the different groups for each character revealed significant differences (p <0.001) between the 5 clusters for all the 11 considered characters. The characteristics of each cluster are presented in Table 9. Indeed, the cluster I had high pod width (PWi), stem thickness (StT) and 100-seed weight (100SW) accessions and in addition number of seeds per pod (SP) beyond the mean. The cluster II had accessions of 100-seed weight (100SW) similar to the ones of the cluster I while the plant height (PlHe), stem thickness (StT), pod length (PL) and the number of seeds per pod (SP) are very low. The cluster III grouped accessions with maximum number of pods per plant (PPl), number of seeds per pod (SP) and in addition to high yielding and rapid maturing but the plants have the weakness of being tall. The cluster IV and the cluster V grouped the accessions which were late maturing.

Distances between clusters

Inter clusters Euclidian distances varied from 60.48 to 519.79. The highest inter cluster distance (60.48) was

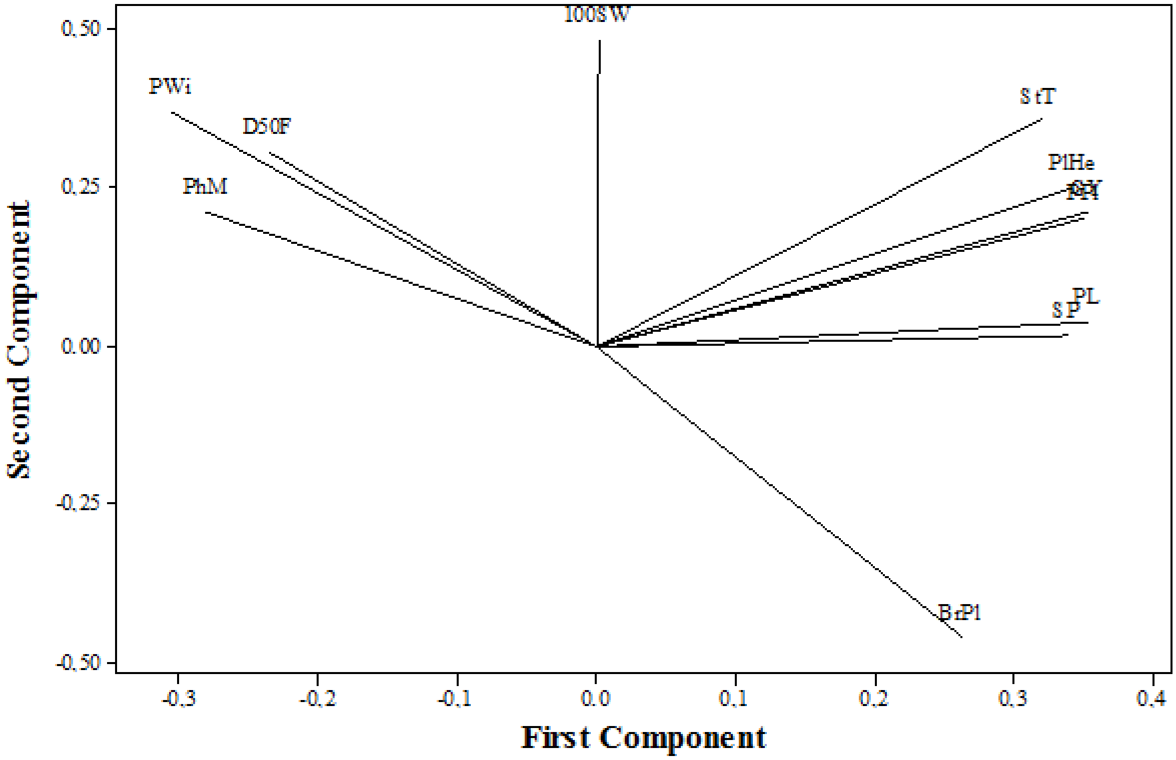

Figure 3. Projection of 11 quantitative characters on the first two components (axis 1 and axis 2) of the PCA.

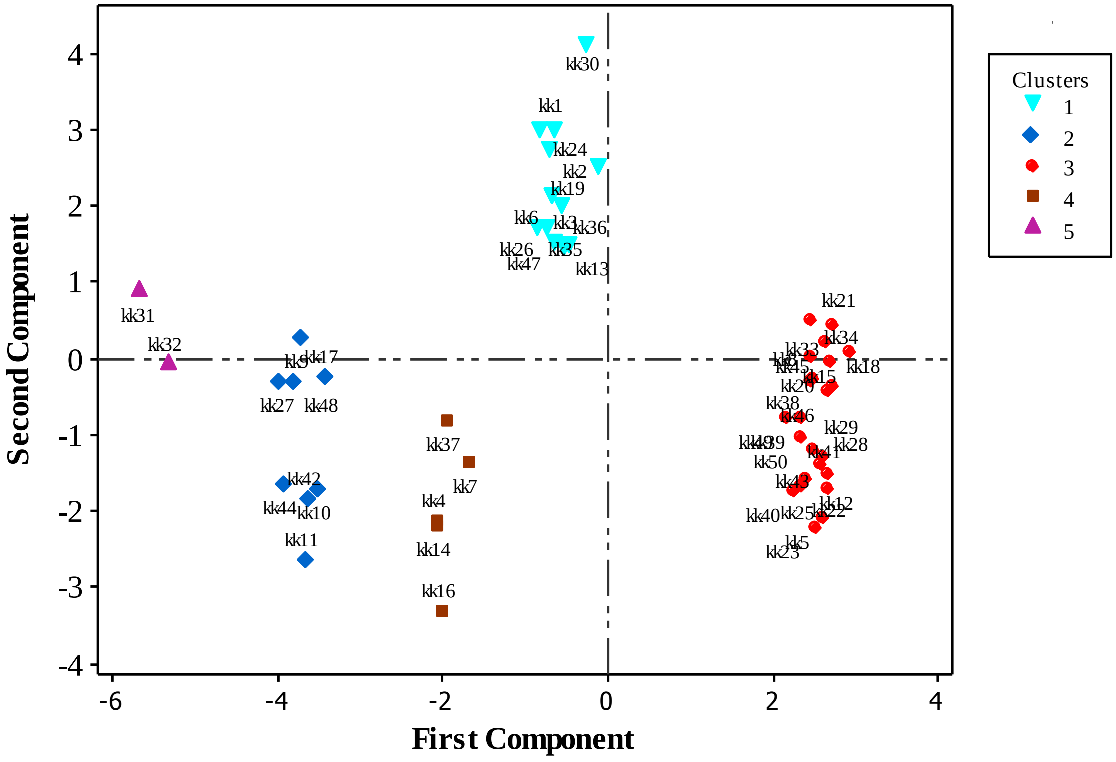

Figure 4. Projection of 50 pigeonpea accessions on the 2 first axes of PCA based on 11 quantitative variables.

Table 9. Comparison of the means of each variable between the five clusters using ANOVA one way and Turkey test.

Variable	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5
PlHe	3.20±0.08^b	1.95±0.03^d	3.30±0.03^a	2.60±0.10^c	1.87±0.01^d
StT	61.70±0.96^a	30.50±0.06^c	60.10±2.28^a	35.42±0.03^b	26.30±0.14^b
BrP	17.75±0.92^d	29.05±0.72^c	44.80±0.77^a	40.64±0.12^b	5.25±0.35^e
PL	61.00±0.94^b	42.50±0.39^d	70.00±0.68^a	60.76±0.20^b	50.50±0.07^c
PWi	8.10±0.03^a	8.00±0.05^b	3.60±0.06^e	5.32±0.05^d	7.12±0.03^c
PPl	1488.90±3.36^b	570.15±0.10^c	1855.20±22.76^a	329.86±0.66^d	135.20±0.85^e
SP	5.05±0.11^c	3.50±0.06^e	5.80±0.03^a	5.21±0.02^b	4.50±0.00^d
GY	4.20±0.15^b	1.86±0.02^d	4.60±0.08^a	2.86±0.03^c	0.57±0.03^e
100SW	12.40±0.06^a	11.70±0.04^a	10.60±0.78^b	7.57±0.04^d	8.97±0.01^c
D50F	165.00±1.01^c	124.58±0.88^d	110.50±0.57^e	174.60±0.21^b	184.63±0.53^a
PhM	190.00±4.77^c	173.00±4.45^d	157.40±0.61^e	199.75±0.10^b	227.88±0.18^a

PlHe: Plant height; StT: Stem thickness; PrBr: Number of primary branches; SeBr: Number of secondary branches; PL: Pod length; PWi: Pod width; PPl: Number of pods per plant; SP: Number of seeds per pod; GY: Grain yield; 100SW: 100-seed weight; D50F: Days of 50% flowering; PhM: Physiological maturity; Averages that have no common letters are statistically different ($p < 0.05$).

Table 10. Inter clusters Euclidian distances.

Cluster	Cluster 1	Cluster 2	Cluster 3	Cluster 4	Cluster 5
Cluster 1	0				
Cluster 2	277.57	0			
Cluster 3	112.43	387.73	0		
Cluster 4	349.65	74.76	460.56	0	
Cluster 5	408.53	133.64	519.79	60.48	0

observed between the cluster III and cluster V, followed by cluster III and the cluster IV (460.56), cluster I and cluster V (408.53), cluster II and cluster III (387.73), cluster I and cluster V (349.65). The lowest inter cluster distance was between cluster IV and cluster V (60.48) (Table 10).

DISCUSSION

Classification of seeds based on their morphological characteristics is the main criteria in folk taxonomy (Akohoue et al., 2018). In the present study of pigeonpea landraces grown in Benin, a real link has been observed between seed classification based on its morphological characteristics and those using morphological qualitative characteristics by grouping accessions in a similar way. This suggests that the morphological characteristics of seeds are important in the evaluation of pigeonpea diversity (Muniswamy et al., 2014). Similar observations have been made on characterization of other legumes such as common bean (Loko et al., 2018), cowpea (Gbaguidi et al., 2013), and Kersting groundnut (Assogba et al., 2015; Akohoue et al., 2018). This confirm that folk taxonomy is not obsolete and can remain for a long time an important preliminary step in the characterization of cultivated genetic resources for further researches.

Our study revealed that seed colour was the highest polymorphic trait. Similar result was found on pigeonpea characterization by Upadhyaya et al. (2007) in Kenya but contrary to those of Manyasa et al. (2008) in Tanzania. This difference can be explained by the fact that the accessions are of different origin. Cream colour and oval-shaped seeds were found to be dominant among pigeonpea landrace grown in Benin. This suggests that landraces with the mentioned traits have been selected by farmers for a long period of time, because of their acceptability by consumers who constitute a key link in the value chain of cultivated genetic resources. Similar observation on seed colour was made on pigeonpea grown in Tanzania (Manyasa et al., 2008; Rao et al., 2012; Kimaro et al., 2017) and Malawi (Rao et al., 2012). This preference for cream seed colour was also observed on other legumes such as Kersting groundnut (Assogba et al., 2015). These characteristics can therefore be considered as varietal preference criteria and should be taken into account by any breeding program of pigeonpea genetic resources in Benin. Majority of pigeonpea landraces showed a strong tendency to semi-spreading growth habit, lanceolate leaflet shape, light yellow base flower colour, and plain seed colour pattern. Similar results have already been reported in the morphological variability of Tanzanian pigeonpea germplasm (Manyasa et al., 2008) and world-wide collection (Rupika and Bapu, 2014). Thus, in spite of the influence of environmental factors, qualitative variables can be used to characterize pigeonpea genetic resources.

Analysis of the genetic characterization of pigeonpea collection based on qualitative characteristics revealed that according to their local names, accessions named differently were grouped into the same morphological class. For instance, landraces kk9 called *Otini tchofiti* (Holly sociolinguistic group), kk10 called *Wlétchivé kloui* (Adja sociolinguistic group), kk11 called *Otili founfoun lakoun* (Fon sociolinguistic group) and, kk17 called *Klouékoun wlanwlan* (Adja sociolinguistic group) grouped in the morphological class C5 on the one hand, and kk35 called *Carder ekloui* (Adja sociolinguistic group), kk36 called *Kloué* (Adja sociolinguistic group) and kk48 called *Klouékoun wéwé* (Fon sociolinguistic group) grouped in the morphological class C1 on the other hand suggests the existence of duplicates in the collection. This fact is not surprising since in the folk nomenclature, the same cultivar through the villages can be designated by different names, which constitute a bias to the estimation of diversity (Agre et al., 2015; Loko et al., 2018). As the identification of duplicates is becoming a priority for genebank managers, molecular genetic characterization would be an efficient approach to discriminate among collection of pigeonpea germplasm (Le clerc et al., 2005; Rana et al., 2015) in order to establish equivalences of names between cultivars (Gbaguidi et al., 2013), but also to reduce the cost of conservation (Horna et al., 2010).

Analysis of the quantitative data showed high level of variation among the 50 accessions with regards to branches per plant, number of pods per plant, pod width and grain yield. This finding suggest the existence of genetic diversity in the pigeonpea landraces grown in Southern and Central parts, which can offer opportunities for genetic improvement in component traits through selection (Pal et al., 2018).

The average grain yield, in our collection (3.73 tonnes/ha) was higher than those obtained in similar studies on pigeonpea (Mergeai et al., 2001; Atta et al., 2008). However, our finding is similar to those observed by Ojwang et al. (2016) and confirm the fact that pigeonpea grain yield can reach up to 5 tons/ha under optimum environmental conditions (Van Der Maesen, 2006) and considering the influence of the environment on certain yields components (Chalak et al., 2018). The average number of seeds per pod estimated at 4.52 was lower than those observed by Kundy et al. (2015). However this number is higher than those observed by Muniswamy et al. (2014) on pigeonpea in India. According to Choudary et al. (2011), the physiological maturity of the cultivars observed in the present study reveal the existence of cultivars with medium and late physiological maturity day.

The correlation analysis of quantitative data revealed strong positive correlation between days to 50% flowering and physiological maturity. Similar results were also reported by Singh et al. (2016); Meena et al. (2017) and Pal et al. (2018) for physiological maturity, on pigeonpea.

These results suggested possibility of indirect selection in correlated traits (Silva et al., 2016) *viz.,* days to 50% flowering cannot be prioritized in selection without effects on physiological maturity. Moreover, the positive significant association between grain yield and plant height, number of branches per plant, pod length, number of pods per plant and number of seeds per pod indicates that these traits are important yield contributing traits in pigeonpea. Thus, should be put into consideration when selecting for yield potential (Ojwang et al., 2016). However, strong negative correlation was observed between physiological maturity and grain yield. Similar finding was observed on pigeonpea in Kenya by Ojwang et al. (2016). This negative correlation between grain yield and physiological maturity should be explained by the lack of enough time by plants to accumulate biomass (Vange and Egbe, 2009; Cheboi et al., 2016) which suggests the presence in our pigeonpea collection of some accessions with short grain filling period. So direct selection for long grain filling periods may increase yield for pigeonpea in Benin. Also high temperatures, low rainfall and high pest infestations constituted such as many factors which involve flower abortion involving low number of pods per plant and 100-seed weight thus lowering the grain yield. Moreover, grain yield is a complex character which is highly influenced by the environment and is the result of interrelationships of its various yield components (Grafius, 1960). Thereby, the negative significant correlation exhibited between plant height, number of branches per plant and number of pods per plants with physiological maturity, implies that plants in our pigeonpea collection mature early and justify the fact that the lack of enough time by plants to accumulate biomass could have been a result of negative correlation observed between physiological maturity and grain yield rather than abiotic (high temperatures and low rainfall) and biotic stress (pest infestations).

This study allowed grouping the 50 accessions into 12 morphotype according to the seed characteristics while the qualitative variables grouped them in 11 morphological types and the Principal Component Analysis grouped them into five clusters. These findings suggested that both qualitative variables and quantitative variables data can reveal diversity providing different but complementary information.

Our results revealed that clustering pattern of the pigeonpea accessions from different origin were frequently present in same cluster. Thus, there was no clear relationship between accessions and geographical diversity. This could be attributed to free exchange of materials that may have overlapped in the previous diversity distribution pattern of the domesticated species (Jaradat and Shahid, 2006; Aghaee et al., 2010). These findings suggest that geographical isolation may not be the only factor causing genetic diversity (Rekha et al., 2011). Therefore, for any hybridization programs in Benin, the choice of suitable diverse parents based on genetic divergence analysis would be more fruitful than the choice based on the geographical distances.

Considering the mean performance for different earliness and yielding traits, the promising genotypes that can be used as parents in hybridization program are those of cluster 3. The high variation of inter clusters Euclidian distances observed in the present study indicated enormous diversity among the genotypes. The highest inter cluster distance was observed between the cluster III and the cluster V suggesting that accessions from these clusters were too much genetically different. However the lowest inter cluster distance between the cluster IV and the cluster V indicated the closer relationship among the genotypes between these clusters. Selection of genotypes from these clusters may not be desirable to get higher yield benefits (Muniswamy et al., 2014; Rupika and Bapu, 2014).

Conclusion

Despite the high diversity in terms of qualitative and quantitative traits, from 23 accessions, kk5 (*Ekloui*), kk8 (*Nontchiovi kloui*), kk15 (*Otili founfoun*), kk18 (*Klouékoun wéwé*), kk22 (*Otili*), kk23 (*CA monlikoun*) and kk28 (*Hounkoun wéwé*) were identified in this study. Our results indicated that the higher level of genetic diversity observed within collected accessions will enable efficient utilization and pigeonpea improvement in breeding programs. Further characterization using molecular techniques as well as conservation attention for these local germplasms should be conducted.

CONFLICT OF INTERESTS

The authors have not declared any conflict of interests.

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