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GENETIC DIVERSITY IN *DETARIUM MICROCARPUM* GUILL & PERR (SWEET DATTOCK) AS MEASURED BY AGRO-MORPHOLOGICAL CHARACTERS

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ABSTRACT: *Nine quantitative agro-morphological characters were studied in nine genotypes of Detarium microcarpum to determine the level and patterns of variation among the genotypes at the nursery stage for breeding record. The genotypes were grown at the Nursery of National Centre for Genetic Resources and Biotechnology, Ibadan, Nigeria. Principal Components Analysis (PCA) and Single Linkage Cluster Analysis (SLCA) were employed to analyze the variation in these genotypes. The result showed that there are variations among the genotypes with respect to number of branches (NoB), number of leaves (NoL), germination percentage (G%), internode distance (ID), seedling girth (SG), leaf length (LL) and leaf width (LW). The first principal components axis accounted for 69.86% of total variation observed among the nine genotypes while the first two principal components axes accounted for 86.62%. SLCA further summarized the relationship among the genotypes into four clusters; cluster 1 (BA-T, KW-AL, JG-D, KD-B, KN-S, PL-J), cluster 2 (KB-Y), cluster 3 (OY-I) and cluster 4 (NG-K) with cluster 2, 3, and 4 genotypes as distinct clusters but clusters 3 and 4 are most distinct of all the genotypes. This implies that cluster 3 and cluster 4 can be used as male parents to cross cluster 1 and cluster 2 to improve the characters such as NoB, NoL, G%., ID, SG, LL, and LW.*

KEY WORDS: Genetic diversity; *Detarium microcarpum;* Agro-morphological Characters; Seedlings, PCA, SLCA, Clusters, Genotypes

INTRODUCTION

Many are the plant resources in the wild that are yet to be fully utilized in term of research, among which is *Detarium microcarpum* Guill & Perr. The plant is an underutilized natural resource of savanna origin with various usefulness for the local inhabitants in which it is found.Sweet dattock is a shallow-rooted, deciduous shrub or small tree with a twisted bole and widely spreading crooked branches, growing up to 9 - 10 metres tall [1, 2]
A valuable multipurpose tree, it is widely utilized for food, medicine and various commodities throughout its range [3]. The leaves and roots are commonly sold in local markets for medicinal purposes [3]. Although not cultivated, it is often left growing when forest is cleared for farmland [4]. In spite of various uses sweet dattock can be put into, a setback is highly recognized in its regeneration which is still at the natural level due to its wild state. Plant species of such nature needs to be domesticated to cater for the human population which grows geometrically against the food production of arithmetic progression. But for wild plant to be domesticated, such plants needs to be studied at all stages of growth and development with special interest in genetic make-up of

such plant species among the diversity available to be ascertain of which among the diversity has the trait(s) desired or the trait(s) that can be easily manipulated to achieve such desired traits. It is on this note that [5] reported that genetic diversity was very much important factor for any hybridization program aiming at genetic improvement of yield.

[6] reported that phenotypic variation is positively associated with genetic diversity, yet also depends on environmental factors and the interactions between genotype and environment. Genetic diversity may be derived from breeding populations (either naturally occurring or synthetic), segregating progeny from a cross of selected parental lines, exotic materials that are not adapted to the target environment, wide interspecific crosses, naturally occurring or induced mutations, the introduction of transgenic events, or combinations of these sources.

Morphological technique in measuring genetic diversity is of importance since it deals with vegetative and reproductive form and structure of plant which forms the basis of observable characters in term of germination count, height, number of leaves, leaf area, fruit/seed length, fruit/seed weight, leaf colour, fruit/seed colour, fruit taste et cetera. In encompassing, it can be broadly grouped into quantitative and qualitative breeding. Employing morphological approach as a technique, consideration should be made to document the plant genetic variations in all stages of plant phenology such as seedling stage, juvenile stage, growing stage, reproduction stage and fruiting to maturity stage for both annuals and perennials.

The Plant Species Distribution and Uses

It is commonly known as sweet detar, sweet dattock or tallow tree, an under-utilized leguminous tree that grows naturally in the drier regions of West and Central Africa [7]. Many different vernacular names exist for this species, including the English, sweet dattock or tallow tree, and the French, dankh or petit détar, as well as Abu-laili (in Sudan) or Tamba Dala (in Mali) [8]. In Nigeria, the plant is known as "Taura" by Hausa tribe, in Igbo, it is called "Ofor", Kanuri calls it "Gatapo" while among Yorubas, it is called "Ogbogbo" [9]. *D. microcarpum* is classified as a major African medicinal plant. The roots, stems, bark, leaves and fruits are all used to treat ailments such as tuberculosis, meningitis, itching, syphilis and diarrhea [10, 11, 12, 13]. Isolation of terpenoids and anti-HIV flavans from *D. microcarpum* extracts have been reported by [11].

Morphology

The morphological descriptors that best describe differences between populations are leaf length, endocarp shape, seed shape, pulp thickness, leaf width, leaf area and the number of leaves per plant. Large trees with high basal branches, very wide leaves, long and heavy fruits and heavy seeds are typical of the Sahel zone, while trees with small circumference and fruit with very thick pulp are typical of the northern Sudan zone. Trees from the southern Sudan zone are characterized by long leaves and very large fruit. The number of leaves per tree has been reported to be inversely proportional to the pulp thickness: [9]. A morphological and biochemical characterization of different populations of *Detarium microcarpum* in Southern Mali was made as the first step in a study of the genetic structure of the species. The farmers' criteria for distinguishing types of *Detarium microcarpum* relate especially to bark colour, leaf size and fruit quality [12].

The assessment of the genetic relationships among the trees at morphological level is a fundamental component of crop improvement programmes. This will provide information about genetic diversity, identification of diverse parental combinations to develop segregating progenies with maximum genetic variability for further selection [14].

Sequel to the plant availability in the area and of its greater ethno-botanical values among the locals, it is noteworthy to research into its diversity through morphological approach for future domestication purposes. It is in view of this that the research was staged up to characterize the morphological constituents of *Detarium microcarpum* at seedling stage for proper identification and record keeping on seedling vigours of various accessions of the species to serve as foundation record for breeding towards domestication.

MATERIALS AND METHODS

Sources of Planting Materials

The matured fruits of *Detarium microcarpum* were collected from nine different states in Nigeria across Derived savanna, Guinea savanna, and Sudan savanna. The fruits were bulked according to the location of collection. The genotypes names were coded based on the locations of collection (Table: 1).

Table 1: Derived Names and Sources of *Detarium microcarpum* used for the Study

S/N	GENOTYPES	SOURCES	VEGETATION ZONES
1.	**OY-I**	Igbeti, Oyo State	Derived savanna
2.	**KW-AL**	Kaiama, Kwara State	Derived savanna
3.	**NG-K**	Kainji, Niger State	Guinea savanna
4.	**KB-Y**	Yauri, Kebbi State	Guinea savanna
5.	**PL-J**	Jos, Plateau State	Montane
6.	**KD-B**	Buruku, Kaduna State	Guinea savanna
7.	**KN-S**	Sitti, Kano State	Sudan savanna
8.	**JG-D**	Dutse, Jigawa State	Sudan savanna
9.	**BA-T**	Toro, Bauchi State	Sudan savanna

Source: Field study, 2019

Experimental Site

The seeds were grown at the Nursery of National Centre for Genetic Resources and Biotechnology, Ibadan, Nigeria with point coordinates of Longitude 3.840N and Latitude 7.384E.

Experimental Layout

The experiment was laid-out in a Randomized Complete Block Design with three replicates. 15 seeds of the plant per state were planted in each replicate. Seeds were mechanically scarified to aid uniform germination and planted in polythene bags filled with loamy soil.

Seeds Extraction and Processing

The seeds were extracted by pod breaking which was accomplished by stones manually. The seeds were then picked and cleansed. Broken seeds were discarded.

The unbroken seeds were tested for viability using water floatation method. A clean bowl was filled with clean water. The clean seeds were then poured inside the water in the bowl. Any seed floated was considered non viable while those that wholly or partially immersed in the water was considered viable with living cotyledons. The seeds with the living cotyledons were then picked for planting.

Sowing of Seeds

To avoid bias in emergence of various genotypes of *Detarium microcarpum* under study, a straight stick of 11.9mm diameter was cut, peeled and graduated in millimeter to give a uniform sowing depth for all the seeds. Uniform sowing depth of 15mm was used. The seeds were planted horizontally with their flat sides on the soil surface in black polythene pots of 29cm by 19cm size filled with loamy soil.

Silvicultural Management

The research units were subjected to watering twice a day from the day of planting; early in the morning before sun rise between 6:00a.m and 7:30a.m, and in the evening between the hours of 5:00p.m and 6:30p.m. The water was given to soil saturation point through out the research period with the use of watering can.

Weeds were controlled by rogueing.

Data Collection

8 seedlings per state, per replicate were randomly selected for data collection making a total of 24 seedlings per state. Data were collected on nine agro- morphological characters for a period of twelve weeks. (Table: 2).

Table 2: Agro-morphological Characters Studied and Methods of Measurement

S/N	CHARACTERS STUDIED	METHODS OF MEASUREMENT
1.	Number of Branches.	The branches originated from the main stem were numerically counted. **By Counting**
2.	Germination %	Number of germinated seeds/ Total number of seeds planted x 100 was calculated. This is neither a parameter nor character but derivative. **By Calculation.**
3	Internode distance	The distance from one branch to another on the main stem was measured by graduated ruler. **By Measurement.**
4.	Seedling height	The height of each seedling was taken from the soil surface to the tip of the seedling with the aid of graduated ruler. **By Measurement.**
5.	Seedling girth	Circumference of each seedling was taken from the base of the first leaves with the aid of digital Vernier caliper. **By Measurement.**
6.	Number of leaves	Every leaf on each seedling was counted. **By Counting.**
7.	Leaf length	This was taken by graduated ruler from leaf base to leaf tip. **By Measurement.**
8.	Leaf width	This is the measurement of the widest part of the leaf by graduated ruler. **By Measurement.**
9.	Leaf area	This was taken using modified gravimetric method. **By Weighing and Calculation.**

Source: Field study, 2019

Data Analysis

The mean value for each measured character was computed for the nine genotypes used. ANOVA was carried out using [15] package.

Principal Component Analysis (PCA) was performed to determine the pattern of variation and the percentage contribution of each character to total genetic variation.

Single Linkage Cluster Analysis (SLCA) was carried-out to summarize the relationship among the genotypes.

Morphological Dendrogram was drawn to identify the most distinct genotypes among the nine genotypes.

RESULT

Analysis of Variance

The combined analyses of variance for number of branches, number of leaves, germination percentage, internode distance, seedling height, seedling girth, leaf length, leaf width and leaf area revealed highly significant ($P<0.001$) genotypic variation for all the agro-morphological characters except germination %, internode distance, and leaf area which showed significant ($P<0.01$) variation and seedling height and leaf width that showed non-significant in genotypic variation among the nine genotypes of *Detarium microcarpum*.(Table 3)

Table 3: Mean Squares in the ANOVA of Agro-morphological Characters from Nine Genotypes for *Detarium microcarpum*

SV	DF	NoB	NoL	G%	ID (cm)	SH (cm)	SG (cm)	LL (cm)	LW (cm)	LA (cm^2)
Block	2	0.17	4.04	283.09	0.03	4.64	0.02	0.03	1.55	9.34
Genotype	8	1.07***	578.81***	620.17***	0.13***	9.08	0.19***	0.57***	1.00	55.11**
Error	16	0.06	4.09	96.07	0.02	5.99	0.01	0.03	0.68	14.04

****, *** = Significant at 0.01 and 0.001 probability respectively**

NoB = Number of branches, NoL= Number of leaves, G%= Germination Percent, ID = Internode Distance, SH = Seedling Height, SG = Seedling Girth, LL= Leaf length, LW = Leaf Width and LA = Leaf Area.

Table 4 presents the mean performance of nine genotypes of *Detarium microcarpum* evaluated for nine agro-morphological characters. All genotypes are significantly different in relation to the administered characters except leaf width and germination percentage that show strong non-significant, and non-significant variation respectively among the genotypes.

Table 4: Mean Performance of Agro-morphological Characters from Nine Genotypes of *Detarium microcarpum*

Genotype	NoB	NOL	G%	ID (cm)	SH (cm)	SG (cm)	LL (cm)	LW cm	LA (cm^2)
OY-I	5.50^{a}	55.28^{a}	42.22^{b}	2.02^{b}	24.02^{d}	2.58^{a}	6.28^{a}	4.01^{a}	20.79^{cd}
KN-S	4.33^{b}	14.08^{bc}	88.89^{a}	2.35^{bc}	26.28^{ab}	1.94^{cd}	5.41^{bc}	2.93^{a}	31.91^{a}
NG-K	4.25^{bc}	16.42^{b}	82.22^{a}	2.37^{bc}	27.24^{ab}	2.19^{b}	6.02^{a}	4.33^{a}	18.04^{d}
KD-B	3.96^{bcd}	14.75^{bc}	86.67^{a}	2.61^{ab}	26.23^{ab}	1.95^{cd}	5.26^{bcd}	2.86^{a}	25.65^{abc}
BA-T	3.87^{bcde}	15.08^{bc}	82.22^{a}	2.57^{ab}	28.40^{a}	1.76^{d}	5.12^{cd}	2.88^{a}	24.32^{bcd}
PL-J	3.87^{bcde}	11.58^{c}	79.99^{a}	2.59^{ab}	25.83^{ab}	2.00^{ab}	5.56^{b}	2.99^{a}	23.79^{bcd}
KW-AL	3.79^{ced}	13.17^{bc}	86.67^{a}	2.50^{ab}	28.11^{a}	1.75^{d}	5.11^{cd}	2.86^{a}	27.75^{abc}
KB-Y	3.75^{ed}	11.58^{c}	82.22^{a}	2.24^{cd}	23.28^{cd}	2.04^{bc}	4.98^{d}	2.81^{a}	28.66^{ab}
JG-D	3.42^{e}	14.46^{bc}	86.67^{a}	2.65^{a}	27.36^{ab}	1.94^{cd}	5.32^{bcd}	2.83^{a}	28.39^{ab}

NoB = Number of branches, NoL= Number of leaves, G%= Germination Percent, ID = Internode Distance, SH = Seedling Height, SG = Seedling Girth, LL= Leaf length, LW = Leaf Width and LA = Leaf Area.
Means with the same letters in a column are not significantly different

Principal Component Analysis

The two principal components accounted for 86.62% of the total variance, with the first principal component taking 69.86%. The relative Eigen values was high (6.29) for axis 1 and low (1.5) for axis 2. The first principal component was mostly correlated with the number of branches, number of leaves, germination percentage, internode distance, seedling girth, leaf length and leaf width. The characters that were mostly correlated with the second principal component were seedling girth and leaf area (Table 5).

Table 5: Principal Component Analysis of Some Major Characteristics of the First Two Axes in *Detarium microcarpum* Genotypes

CHARACTERS STUDIED	COMPONENT AXES	
	AXIS 1	AXIS 2
Number of branch	0.37	-.07
Number of leaf	0.36	-.09
Germination %	-3.7	0.11
Internode distance	-3.3	0.34
Seedling height	-2.1	0.59
Seedling girth	0.38	-0.7
Leaf length	0.36	0.28
Leaf width	0.33	0.38
Leaf area	-2.6	-5.4
Eigen values	6.29	1.51
Total variation %	69.86	16.76
Cumulative variation %	69.86	86.62

Correlation Coefficient Analysis

Table 6 shows the correlation coefficients of the nine agro-morphological characters that were used in characterizing the nine genotypes of *Detarium microcarpum* among the nine genotypes of *Detarium microcarpum.* Positive genotypic correlation occurred between germination percentage and leaf area. Also, the correlation matrix showed the strong negative genotypic correlation between leaf area and leaf length. However, both internode distance and seedling height had positive but non-significant genotypic correlation with leaf area.

Table 6: Correlation Coefficient among Seedling Characters of Nine *Detarium microcarpum* Genotypes

	NoL	**G%**	**ID**	**SH**	**SG**	**LL**	**LW**	**LA**
NoB	0.81**	-0.77**	-0.67*	-0.43*	0.77**	0.72**	0.52**	-0.33
NoL		-0.79**	-0.62**	-0.21	0.77**	0.69**	0.32	-0.36
G%			0.59**	0.47*	-0.77**	-0.64**	-0.56***	0.43*
ID				0.42*	-0.63**	-0.39*	-0.24	0.16
SH					-0.50**	-0.24	-0.33	0.08
SG						0.80**	0.63**	-0.41*
LL							0.63**	-0.63**
LW								-0.31

***, ** =Significant at 0.05 and 0.01 probability levels respectively.**

NoB = Number of branches, NoL= Number of leaves, G%= Germination Percent, ID = Internode Distance, SH = Seedling Height, SG = Seedling Girth, LL= Leaf length, LW = Leaf Width and LA = Leaf Area.

Single Linkage Cluster Analysis

Figure 1 shows the single linkage cluster analysis which illustrated the relationship based on the characters evaluated among the nine genotypes under study. The analysis put up the clustering distribution into four. Cluster 1 includes BA-T, KW-AL, JG-D, KD-B, KN-S and PL-J, Cluster 2 comprises KB-Y, Cluster 3 contains OY-1 and Cluster 4, NG-K. All genotypes in cluster 1 are similarly significant while cluster 2, 3 and 4 are widely distributed with KB-Y, OY-I and NG-K showed as distinct genotypes of *Detarium microcarpum.*

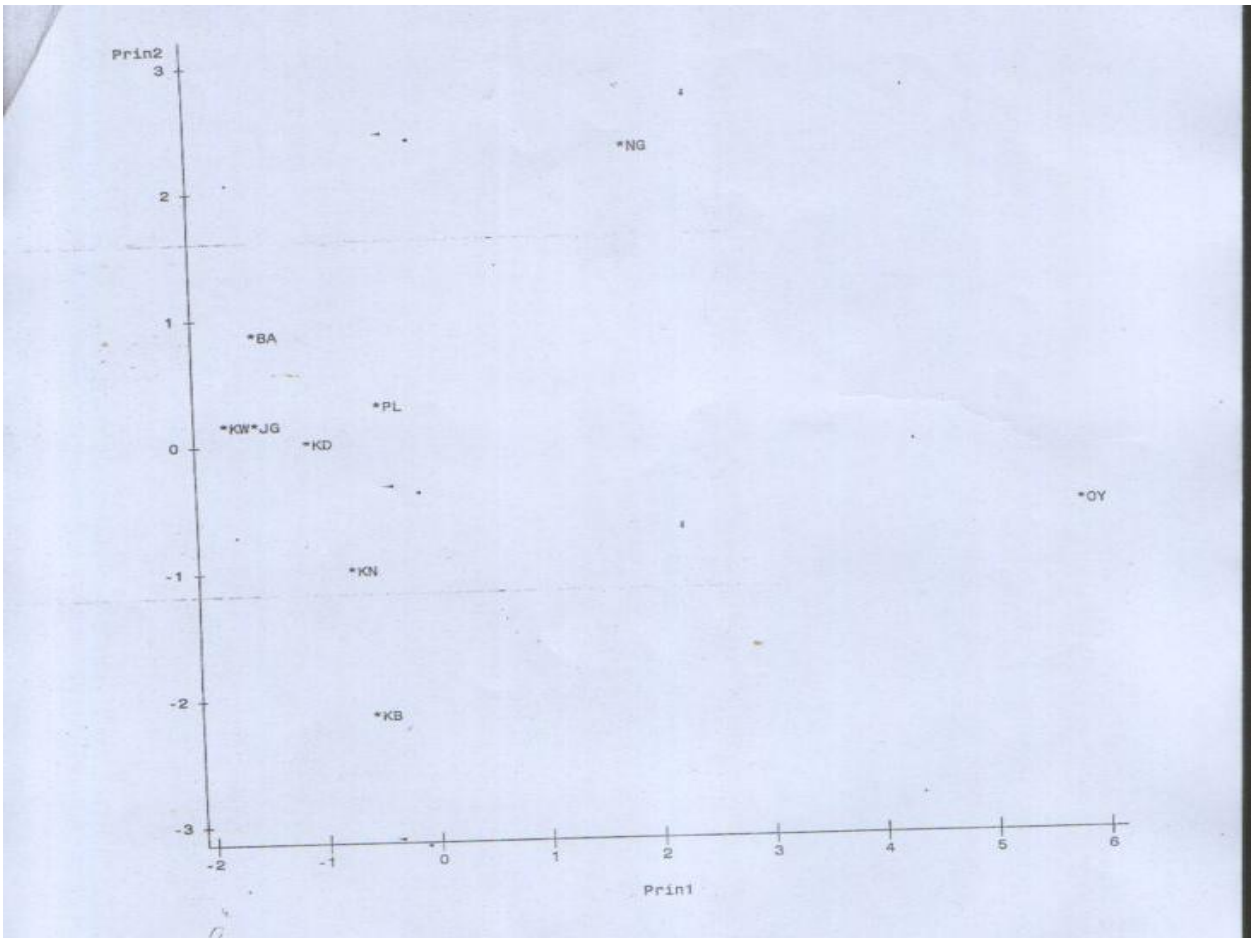

Figure 1: Single Linkage Cluster Analysis (SLCA) of Nine Genotypes of *Detarium microcarpum*

Morphological Dendrogram

Dendrogram originated from (SLCA) showed six genotypes forming cluster 1; BA-T, KW-AL, JG-D, KD-B, KN-S having no significant variation, but, KB-Y (cluster 2) slightly varies from cluster 1 (six genotypes), NG-K (cluster 4) widely significant from cluster 1and OY-I (cluster 3) as most distinct among the nine genotypes. (Figure 2)

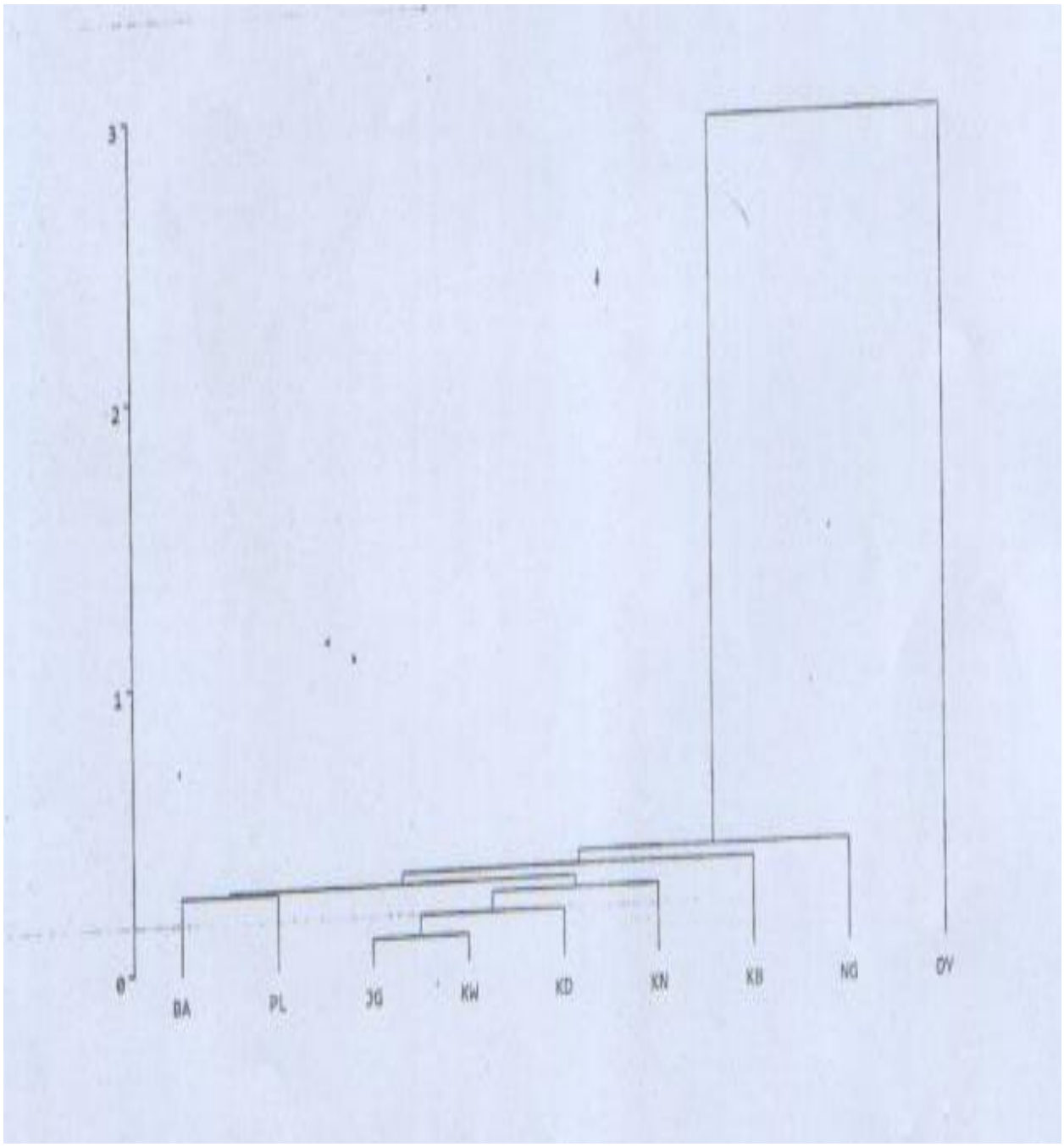

Figure 2: Dendrogram illustrating relationship among the Nine Genotypes of *Detarium microcarpum*

DISCUSSION

This study assessed the genetic diversity in *Detarium microcarpum* Guill and Perr as measured by nine agro-morphological characters at seedling stage. Genetic variation was observed based on the characters evaluated except seedling height and leaf width. The result of the principal component

analysis showed that different characters contributed differently to the total variation in the Detarium genotypes, as indicated by Eigen value as well as their weight and loading in different principal axes. The first principal component that accounted for the highest proportion of the total variation indicated the contribution of NoB, NoL, Germ, ID, SG, LL, and LW. If selection was to be made between the cluster groups for a future breeding exercise, these characters should be given high priorities.

The cluster analysis had singular efficacy and ability to identify crop genotypes with the highest level of similarity through the Dendrogram generated [11]. Principal Component Analysis (PCA) is a descriptive method which shows the pattern of co-variation of characters among individual [13]. [18] considered PCA as a powerful technique for data reduction as it removes interrelationships among the components. The results reported by various researchers showed the multivariate analysis as a valid system to deal with germplasm collection [19]. [20] studied numerical analysis of variation among Nigerian accessions of "Egusi" melon (*Citrullus lanatus*) (Thunb.) Matsum and Nakai, using the cluster and principal component analysis. [21] determined the selection of parent for improvement of restorer line in rice (*Oryza sativa L.*) through the use of cluster and principal component analysis. The morphological dendrogram generated from the similarity or distance matrices had provided an overall pattern of variation as well as the degree of relationship among genotypes.

CONCLUSION

From the result, it was concluded that there were variations among the genotypes of *Detarium microcarpum* collected for the research.
Four clusters were formed morphologically among the genotypes of sweet dattock considered with Cluster 1 having six genotypes closely related, Cluster 2 having one, Cluster 3 having one and Cluster 4 with also one genotype.
Genotypes KB-Y, OY-I and NG-K were shown to be of greater divergence for the characters that would offer a good scope to develop the plant breeding programme of *Detarium microcarpum.*

Recommendation

Genotypes KB-Y, OY-I and NG-K were identified as having greater diversity based on the evaluated characters and therefore recommended as male parents for breeding improvement of the plant.
Parents for breeding programme should be taken from the different clusters and not within the same cluster to achieve high level of variability.
Cluster 1 genotypes due to closeness in diversity should be taken as female parents against other genotypes especially of clusters 3 and 4 to increase the pool of diversity among the groups of genotypes.
The results from this study should be added to the wealth of information on the study of *Detarium microcarpum.* Molecular characterization is therefore recommended as a follow-up to uphold the variation pattern among Nigerian *Detarium microcarpum* towards the domestication of the species.

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APPENDIX

APPENDIX 1: Aerial View of *Detarium microcarpum* Tree showing the Fruits, Branches and Leaves Arrangement

APPENDIX 2: A *Detarium microcarpum* Trunk showing the kind of bark possessed.

APPENDIX 3: A Photograph of Matured Fruits of *Detarium microcarpum*

APPENDIX 4: Extracted Seeds of *Detarium microcarpum*

APPENDIX 5: A Photograph of *Detarium microcarpum* Seedlings