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RESEARCH ARTICLE

Morpho-Genetic Characterization of *Abelmoschus* Moench. Accessions

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ABSTRACT

This study was carried out to assess the morphological and genetic variability of seven accessions of *Abelmoschus* L. species using morphometric analysis and ribulose bisphosphate carboxylase large chain (RBCL) molecular markers. Using a completely randomized block design, seeds of the okra accessions were planted with three replicates each. During the developmental stage, the morphological features of the accessions were observed and recorded according to the standard descriptor for the crop. Morphologically, all the accessions exhibited a degree of similarities, albeit, at maturity, the leaf, plant height, leaf colour, and leaf shape became distinct. A cluster of the phenotypic characterization was observed at a 3.74 level of coefficient of similarity with two distinct clusters, which were predicted to be *A. esculentus* and *A. caillei*. The percentage variance of the two principal components was 55.12% and 22.69% with corresponding Eigenvalues of 4.11 and 1.69, respectively. Results of the RBCL analysis revealed genetic variability at a 0.80 level of coefficient of similarity. Two distinguishable clusters were observed. Both morphometric and genotyping results suggest that variations exist among and within the seven accessions. In conclusion, there is a need to frequently evaluate plant genetic resources held in gene banks as they may not reflect the whole range of diversity inherent in the species.

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Introduction

Abelmoschus Moench. species is an important traditional vegetable crop in the tropical, subtropical, and Mediterranean regions of the world (Staub et al., 1997; Bisht & Bhat, 2006; Ogwu & Osawaru, 2022). It is commonly known as okra and the Angiosperm Phylogeny Group (APG, 2009; Osawaru & Ogwu, 2013) recognized it as a monophyletic group. It is cultivated for its edible fruits, leaves, seeds, floral parts, and woody stem (Bioversity International, 2007; Osawaru et al., 2012). It is one of the oldest cultivated crops, presently grown in many countries of the world (Ariyo, 1993; Oyelade et al., 2003; Ogwu et al., 2016a, 2016b, 2017). There are several species, both cultivated and wild especially within their native range. In West Africa, the species of okra cultivars are of two distinct types, the common okra [*Abelmoschus esculentus* (L.) Moench.] and the West African okra [*Abelmoschus caillei* (A. Chev.) Stevels].

Okra may be classified morphologically using the quantitative and qualitative parameters or descriptors recommended by the International Board of Plant Genetic Resources (IBPGR, 1991) and germination protocol as outlined by Osawaru et al. (2012). These descriptors include plant height, leaf size, leaf colour, stem colour, general aspects of the stem, leaf shape, fruit pubescence, *etc*. The morphological characterization points out the high degree of variations that exist among and between accessions of okra, which further requires evidence using molecular makers to clarify

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(Omonhinmin & Osawaru, 2005; Osawaru et al., 2011, 2014a, 2014b). Several biochemical analyses have proven the presence of various phytochemicals embedded in the species. Diversity classified by phenotypic and morphological characters usually varies with environments and evaluation of traits requires growing the plants to full maturity before identification. Presently, the rapid development of biotechnology has allowed easy analysis of the massive number of loci distributed throughout the genome of the plants to allow for variability distinction and characterization. Molecular markers have also proven to be powerful tools for the assessment of genetic variation and in the elucidation of genetic relationships within and among species (Chakravarthi & Narayaneni, 2006). Molecular markers unlike morphological traits are not disturbed by the environment (Staub et al., 1997). Germplasm characterization, a process that involves recording and compilation of data on important characteristics that distinguish one species from the other and accessions or varieties within species, has been used to enable easy and quick discrimination among phenotypes (Bioversity International, 2007; Das et al., 2012). This process determines the expression of highly heritable characters ranging from morphological or agronomical features to seed proteins or molecular markers. Characterization allows for simple grouping of accessions, developments of core collections, identification of gaps, and retrieval of valuable germplasm for breeding programmes resulting in better insight into the composition of the collection and its genetic diversity (Das et al., 2012).

More so, the characterization of genetic resources is an essential first step in any crop improvement and conservation programme (Adeoluwa & Kehinde, 2011; Ogwu et al., 2014). Characterization and quantification of the germplasm and knowledge of the genetic viability between and among closely related crop varieties are necessary for a cogent use of plant genetic resources (Adeoluwa & Kehinde, 2011). A precise knowledge of genetic diversity among okra germplasm plays a major role in breeding programmes.

Molecular characterization has presently become a more efficient method of distinguishing plant genetic resources. It provides reliable information for assessing, among other factors, the amount of genetic diversity, the structure of diversity in samples and populations (Perera et al., 2000; Shim & Jørgensen, 2000), rates of genetic divergence among populations (Maestri et al., 2002), and the distribution of diversity in populations found in different locations (Perera et al., 2000; Maestri et al., 2002; Figliuolo & Perrino, 2004).This study is set up to investigate variations in the morphological and genetic characteristics of the species and characterize them based on their variation.

Materials and Methods

Study Area

This study was conducted at the experimental garden of the Department of Plant Biology and Biotechnology, University of Benin, Benin City, Edo State, Nigeria [6.40°N 5.61°E]. It is located within the humid tropical rainforest zone of Nigeria and belongs to the Af category of Koppen's climatic classification. The climate includes high rainfall up to 200-300 mm of pattern with peaks in July and September respectively, high temperatures ranging between 20-40 °C, and high atmospheric humidity (Omuta, 1980). The RBCL genotyping was carried out at the Bioscience Laboratory of the International Institute of Tropical Agricultural, Ibadan [7.49°N 3.91°E].

Source of Plant Material

Seven accessions of *Abelmoschus* were obtained from the active collection of the national gene bank of the National Centre for Genetic Resources and Biotechnology (NACGRAB), Ibadan Nigeria (Table 1).

Table 1. Passport data of okra accessions collected from NACGRAB.

Accession	Status	Genus	Source Location	Germplasm Collected
NGB 00297	Landraces	*Abelmoschus*	NACGRAB, Ibadan [7.22°N 3.50°E]	Seeds
NGB 00309	Landraces	*Abelmoschus*	NACGRAB, Ibadan [7.22°N 3.50°E]	Seeds
NGB 00308	Landraces	*Abelmoschus*	NACGRAB, Ibadan [7.22°N 3.50°E]	Seeds
NGB 00302	Landraces	*Abelmoschus*	NACGRAB, Ibadan [7.22°N 3.50°E]	Seeds
NGB 00467	Landraces	*Abelmoschus*	NACGRAB, Ibadan [7.22°N 3.50°E]	Seeds
NGB 00371	Landraces	*Abelmoschus*	NACGRAB, Ibadan [7.22°N 3.50°E]	Seeds
NGB 00387	Landraces	*Abelmoschus*	NACGRAB, Ibadan [7.22°N 3.50°E]	Seeds

Morphological Characterization and Data Collection

After five weeks in the field, qualitative and quantitative morphological data were collected based on the International Board for Plant Genetic Resources (IBPGR, 1991) descriptor list for okra. Data included plant height, leaf length, leaf width, and general growth appearance (*i.e.*, habit) [Table 2]. Leaf shape was characterized according to Charrier (1984) [Figure 1].

Table 2. Qualitative morphological characteristics evaluated in the study and their character state and codes.

S/N	Parameters Measured	Parameter Key	Character State
1	General aspect of the stem	GAS	1-Erect 2-Medium 3-Procumbent
2	Stem colour	STC	1-Green 2-Green with red patches 3-Purple
3	Stem pubescence	STP	1-Glabrous 2-Slight 3-Conspicuous
4	Nature of branching	BRA	1-Orthotropic stem only 2-Medium 3-Strong
5	Leaf shape	LSH	From 1-11 (see Figure 1)
6	Leaf colour	LC	1-Green 2-Green with red veins 3-Red

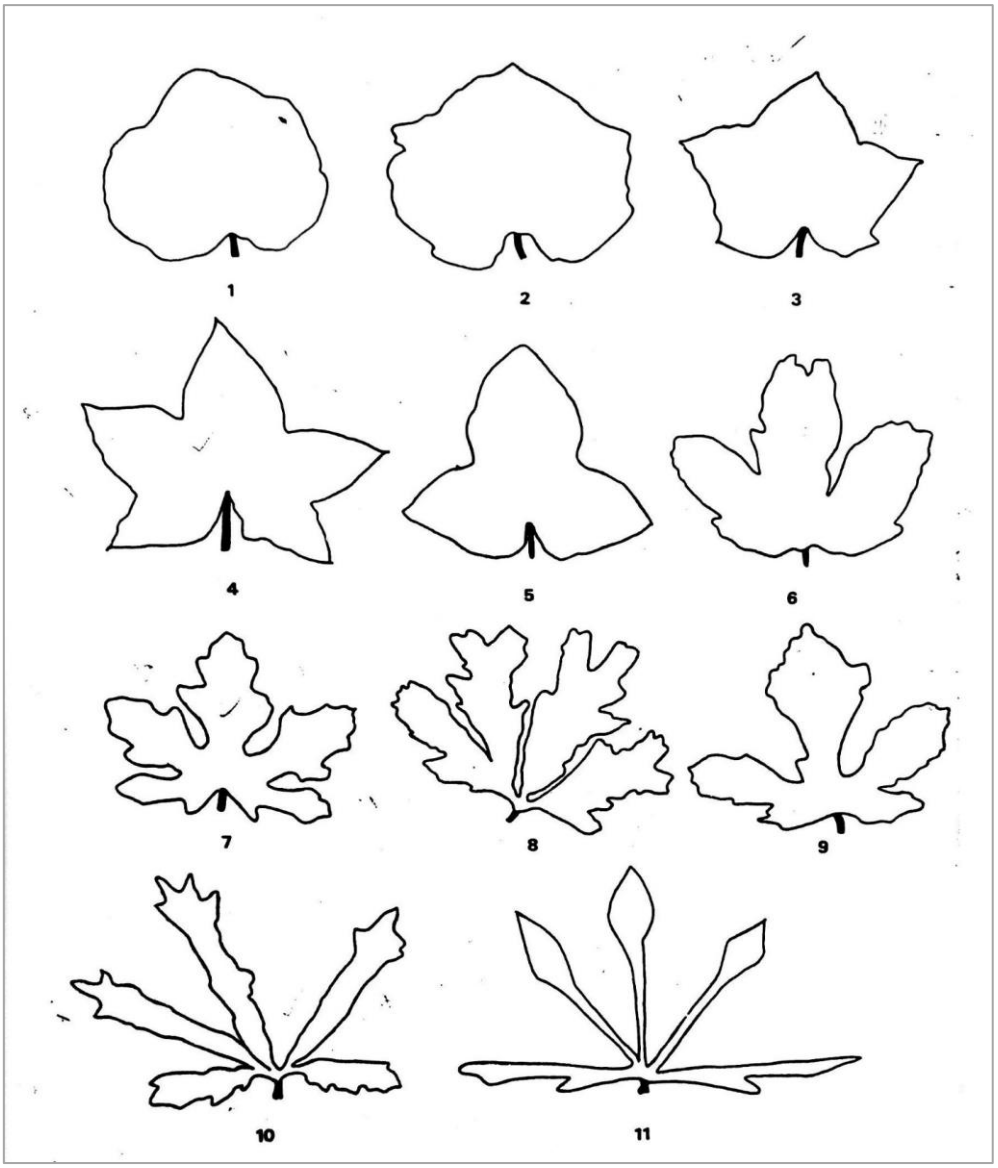

Figure 1. Leaf shapes descriptor key for *Abelmoschus*. Source: Adapted from Charrier (1984).

Ribulose Bisphosphate Carboxylase Large Chain Genotyping

To take into account possible genetic variability within each accession, young leaves of 5 weeks old were genetically analysed using Ribulose bisphosphate carboxylase large chain (RBCL) marker at the Bioscience Laboratory of the International Institute of Tropical Agricultural.

Total genomic DNA was extracted from young leaves (100 mg per accession) of 5 weeks old plants following these procedures. Samples were prepared by putting approximately 100 mg of freeze-dried tissues into an extraction tube. Two steel balls were added each into the tube to enable grinding. The freeze-dried tissue was then ground into a fine powder using Genogrinder-2000. About 450 µl of pre-heated plant extraction buffer was added. The mixture was incubated at 65 °C for 20 min and mixed occasionally by inverting the tubes to homogenize the sample, then the mixture was removed and allowed to cool for 2 mins. 200 µl of ice-cold 5 M Potassium acetate was added and incubated on ice for 20 minutes to precipitate protein. The mixture was Centrifuge at 10000 rpm for 10 mins. The supernatant was transferred into freshly labelled tubes. 450 µl of chloroform isoamylalcohol (24:1) was then added and mixed gently to further precipitate protein and lipids. Again, mixture was centrifuged at 10000 rpm for 10 min and then the supernatant was transferred into freshly labelled tubes. 2/3 volume of ice-cold isopropanol was added to the mixture gently and incubated at -80 °C for 15 mins to precipitate the DNA. The mixture was centrifuged at 10000 rpm for 10 min and the supernatant was decanted until the last drop. Then about 400 µl of 70 % ethanol was added to wash the DNA pellet and centrifuge. The supernatant was decanted until the last drop to remove dry air pellets. 60 µl of ultra-pure water or low salt TE was added to re-suspend the DNA, followed by 2 µl of RNase and then the mixture was incubated at 37 °C for 30-40 minutes. 0.8% agarose gel was prepared to use for checking the DNA quality and removal of RNA. DNA-50 option of the Nanodrop spectrophotometer was used to quantify the DNA concentration. Two universal primers were used for the RBCL-PCR analysis. The PCR mixtures were prepared using 2.5 µl of 10x PCR buffer, 1 µl of 25 mM $MgCl_2$, 1 µl each of forward and reverse primer, 1 µl of DMSO, 2 µl of 2.5 mM dNTPs, 0.1 µl of 5 µl Taq DNA polymerase, and 3 µl of 10 ng/µl DNA. The total reaction volume was made up to 25 µl using 13.4 µl of nuclease-free water. PCR cycling parameters included 9 cycles with an initial denaturing temperature of 94 °C for 5 mins and 15 sec denaturing and afterward an annealing temperature of 65 °C for 20 seconds and finally an extension with 72 °C for 30 seconds. Then 35 cycles with 94 °C for 15 seconds, the annealing temperature of 55 °C for 20 seconds, extension with 72 °C for 30 seconds, final extension at 72 °C for 7 minutes, and then held at 10 °C. After PCR, the amplicons generated were sequenced on ABI3500 Genetic Analyser.

Statistical Analysis

The morphometric analyses, based on measurements of both qualitative and quantitative characters were subjected to multivariate analysis. A matrix was developed based on individual character distribution. Several multivariate approaches were used to compare all the evaluated characters including principal components analysis (PCA), Gower-based non-metric dimensional scaling (NMDS), Gower-based unweighted pair group method with arithmetic mean (UPGMA) cluster analysis, and Pearson (linear) correlation in PAST. Before the analysis, all the characters were normalized using the software's correlation matrix as previously adopted and described by Wahlsteen and Tyler (2019). Pearson's correlation was used to show the relationship between the traits studied. The PCA was used to analyse matrices of several characters and species to get a general overview of the variation in the two groups.

The RBCL matrix generated after sequencing was further analysed using PAST (Palaeontological Statistic) package version 3.24 (Hammer et al., 2019) to compare the similarities and dissimilarities present in the accessions. The principal component analysis was used to analyse matrices of several characters and species to get a general overview of the variation between and among groups. Non-metric dimensional scaling was used to produce a dissimilarity-based index to highlight the taxonomic significance of characters included in a study. Neighbour-joining cluster analysis was used to produce a dendrogram for the accessions.

Results

Reproducible results were obtained from this study for both the RBCL genotyping and morphological characterization. The results from the genetic variability test were complimentary with the morphological characterization. The morphological characteristics were observed in the field weeks after the seeds were propagated. Growth was in stages. Plant height was observed to have a progressive growth pattern in all accessions although accession NGB 00387 was found to have a more rapid growth than the others (Figure 2). Slight similarities were observed between accessions NGB 00297 and NGB 00309; NGB 00302 and NGB 00467; accessions NGB 00371 and NGB 00308 were quite distinct with accession NGB 00308 having a slow growth rate (Figure 2). Leaf length showed a linear growth pattern in all the accessions except NGB 00302, NGB 00371, and NGB 00387, which showed slight similarities in their growth pattern within the first week after germination (Figure 3). The leaf width of the seven accessions showed a distinct growth pattern, as some were much wider than others (Figure 4). Variability among and within the accessions at various stages of their growth was observed. The variations in the qualitative characters of the accessions are presented in Table

3, which further distinguishes them. Leaf shape varied among accessions and this was used to classify the accessions.

The results presented in Figures 5-7 suggest two distinct clusters with each group showing a high level of dissimilarity following the multivariate analysis carried out using data from the morphological traits. Figure 5 shows the principal component analysis for the phenotypic characters of the okra accessions. The two principal components presented represent over 50% of the observed variations within the accessions (Suppl. Table 1). The loadings and scores of the nine principal components are presented in Suppl. Tables 2 and 3, respectively. Figure 6 presents the non-metric dimensional scaling analysis of the accessions. The results support the existence of two clusters based on observed phenotypic dissimilarities within and among the accessions at a stress of 0.103. The loading of each of the NMDS axis is presented in Suppl. Table 4. The R^2 value of axis 1 is 0.851 whereas axis 2 is 0.090. Figure 7 presents the Gower-based UPGMA clustering dendrogram based on the phenotypic traits of the okra accessions. Table 4 presents the correlation matrix of sequence data from the RBCL genotyping of the accessions. Figure 8 shows the principal component analysis of the okra accession prepared from the matrix generated from the RBCL analysis. These results suggested a more in-between and less out of group association from the seven okra accessions with clusters delimited based on okra species into *A. callei* and *A. esculentus*.

Table 3. Qualitative morphological characteristics evaluated in the study and their character state and codes.

		Accessions						
S/N	**Parameters**	**NGB 00297**	**NGB 00309**	**NGB 00308**	**NGB 00302**	**NGB 00467**	**NGB 00371**	**NGB 00387**
1	GAS	Erect	Erect	Erect	Erect	Erect	Erect	Erect
2	STC	Green	Green	Green with red patches	Purple	Green with red patches	Green with red patches	Green with red patches
3	STP	Slight	Slight	Slight	Slight	Slight	Slight	Slight
4	BRA	Orthotropic stem only	Orthotropic stem only	Orthotropic stem only	Strong	Orthotropic stem only	Orthotropic stem only	Orthotropic stem only
5	LSH	Lobed	Cordate	Cordate	Pinnately lobed	Lobed	Cordate	Pinnately lobed
6	LC	Green	Green with red veins	Green with red veins	Green	Green	Green with red veins	Green with red veins

GAS: General aspect of the stem, STC: Stem colour, STP: Stem pubescence, BRA: Nature branching, LSH: Leaf shape, LC: Leaf colour.

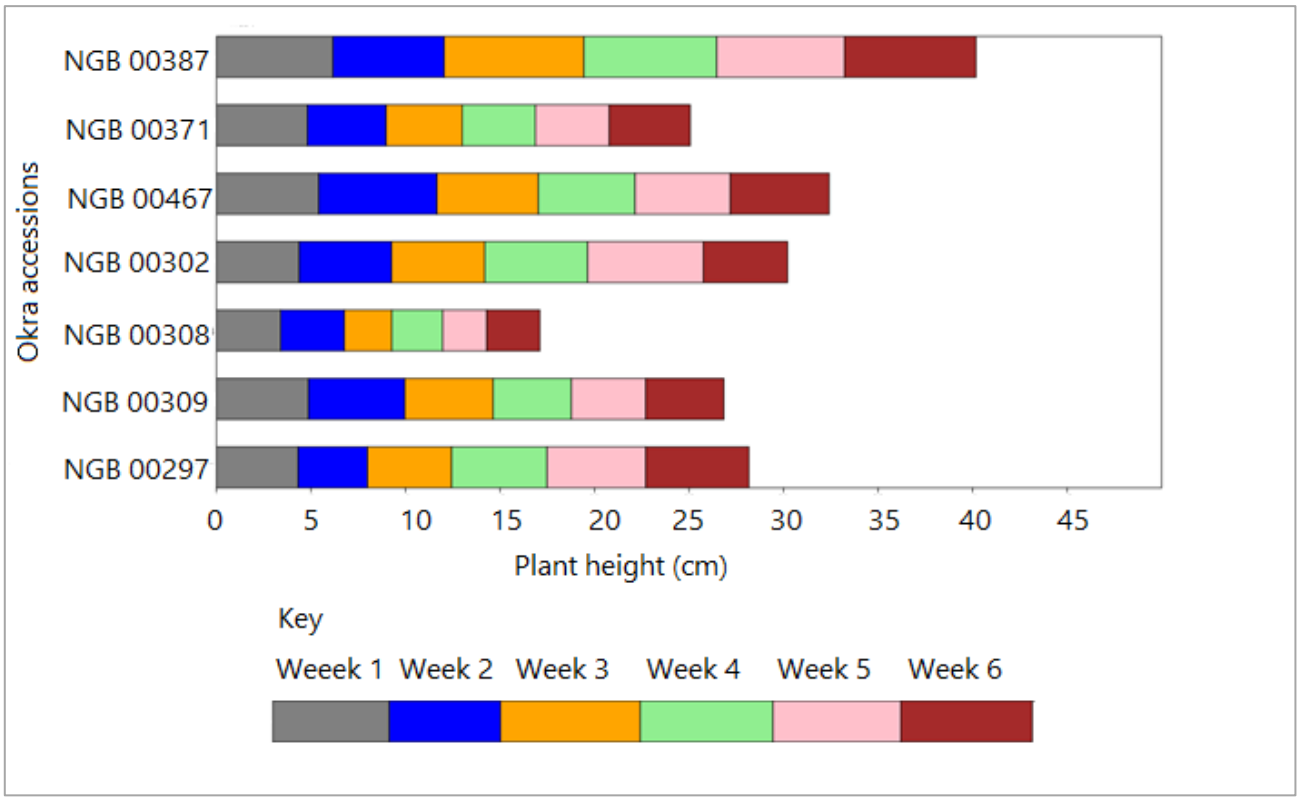

Figure 2. Plant height of okra accessions during the study.

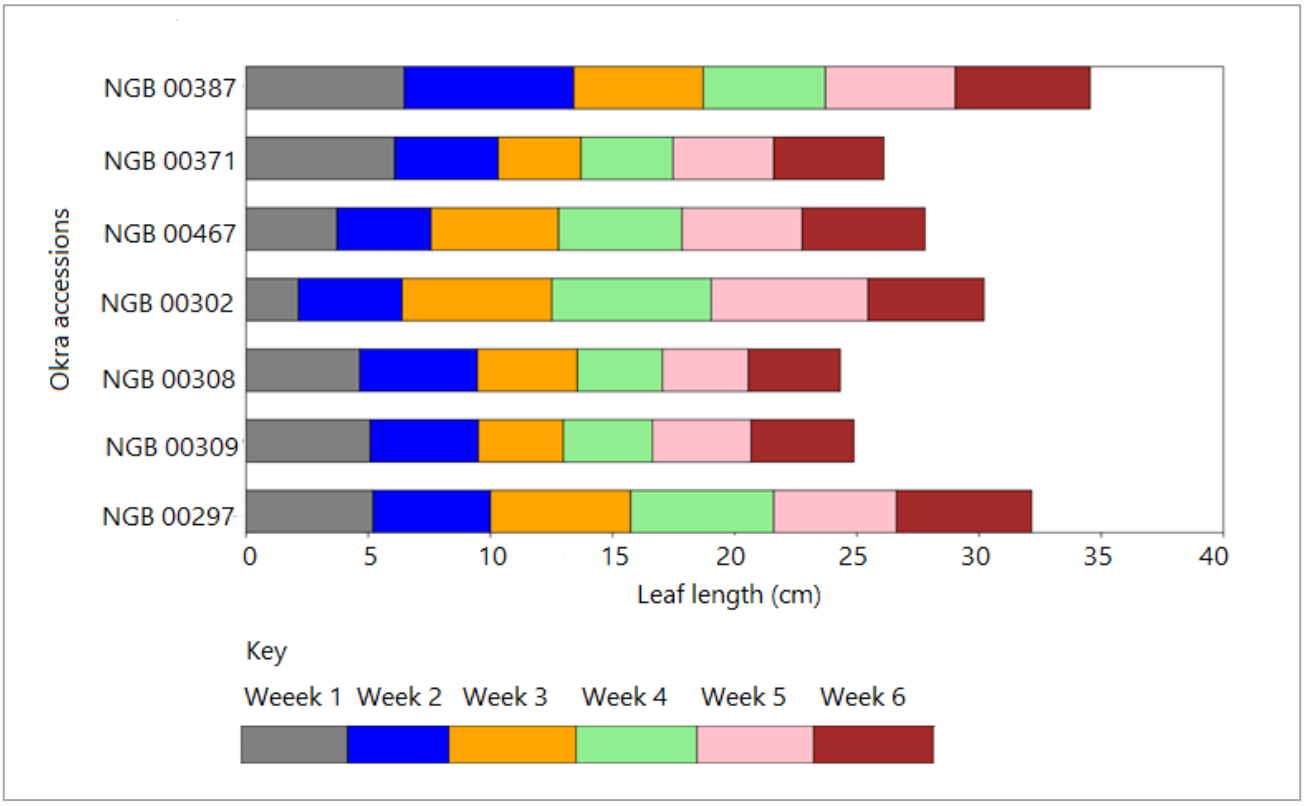

Figure 3. Length of the leaf of *Abelmoschus* accessions during the study.

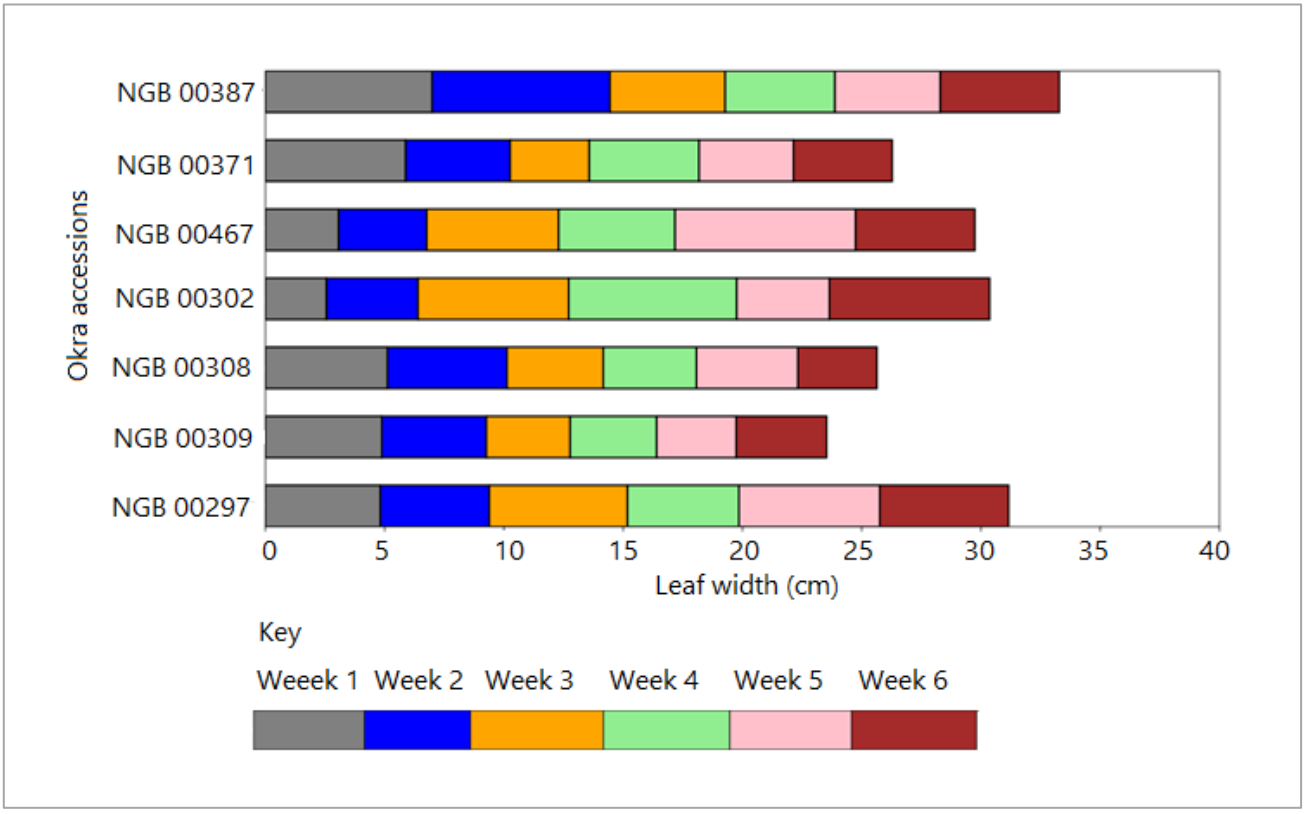

Figure 4. Leaf width of *Abelmoschus* accessions during the study.

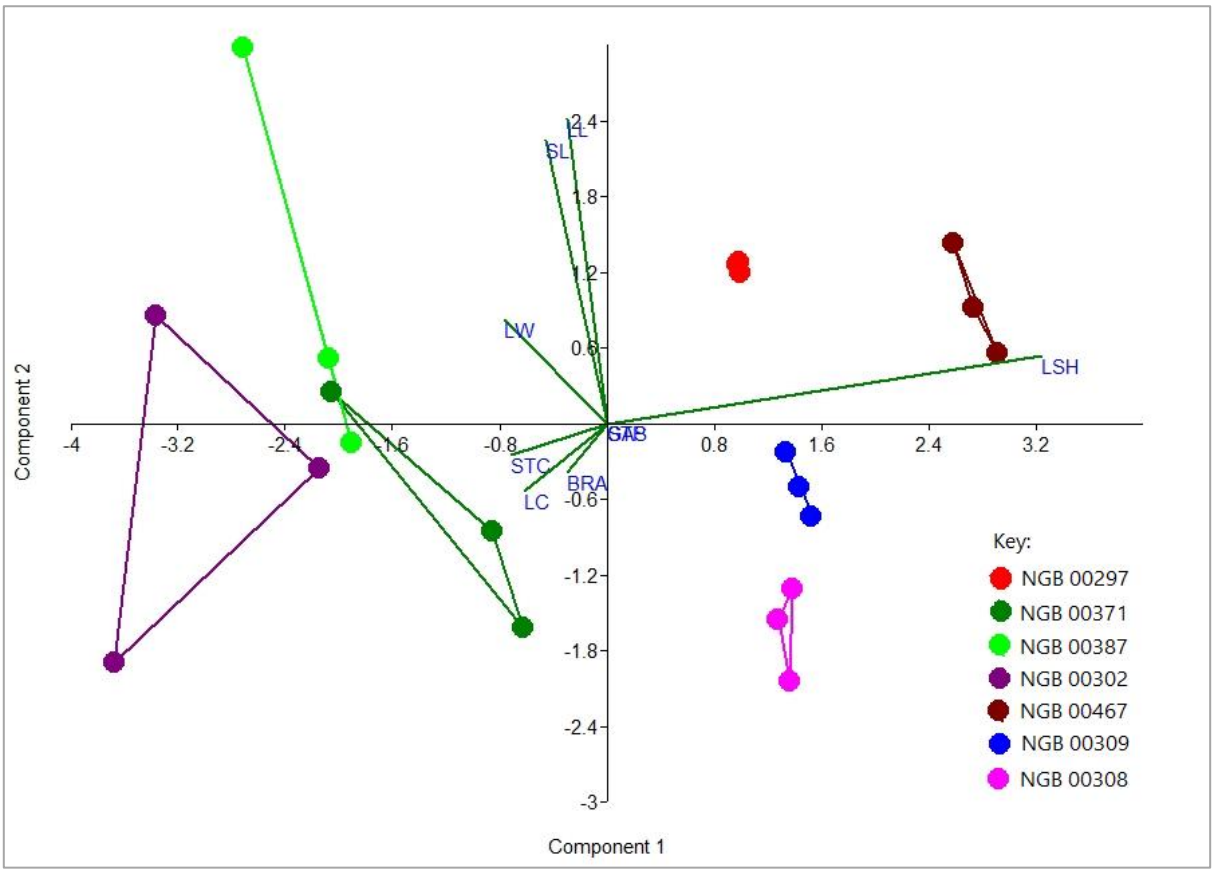

Figure 5. PCA of the phenotypic traits of the okra accessions with Jolliffe cut-off of 0.59963. GAS: General aspect of the stem, STC: Stem colour, STP: Stem pubescence, BRA: Nature branching, LSH: Leaf shape, LC: Leaf colour, LL: Leaf length, LW: Leaf width, SL: Stem length.

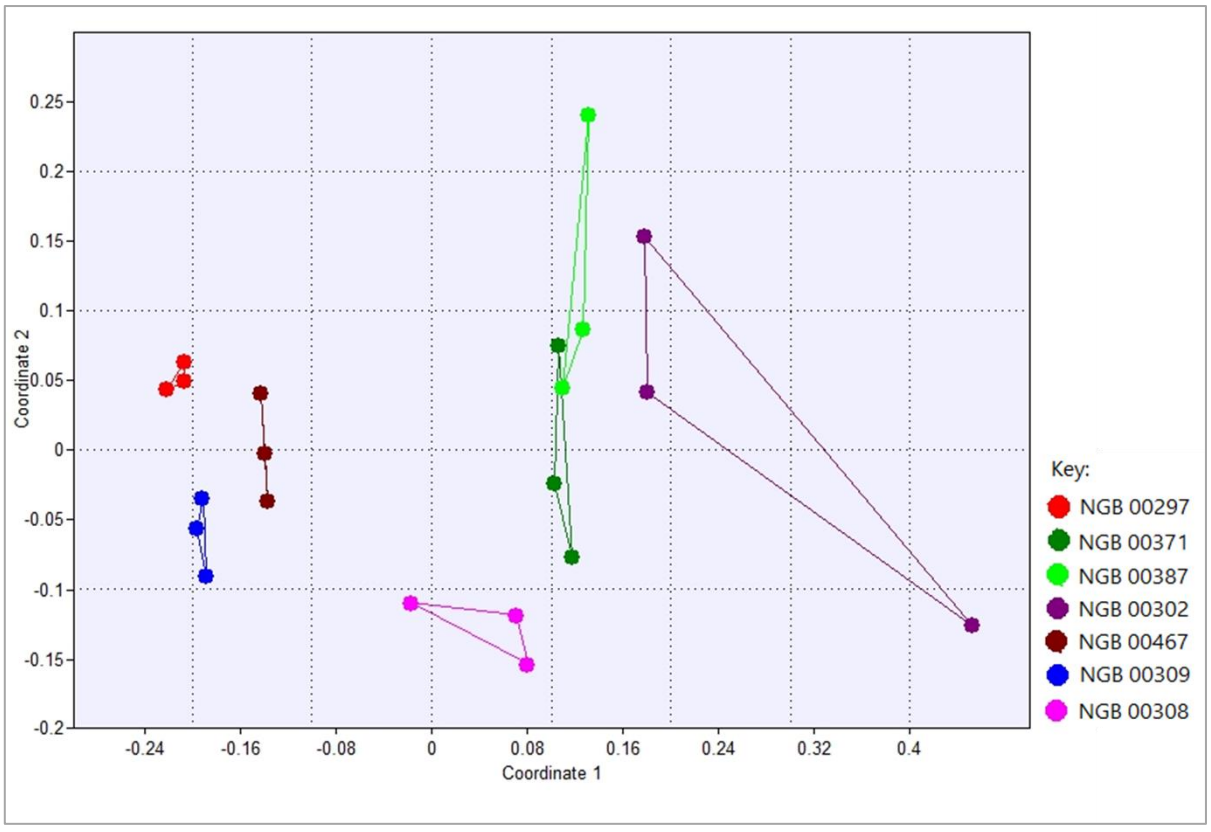

Figure 6. Gower-based NMDS of the phenotypic characters of the okra accessions.

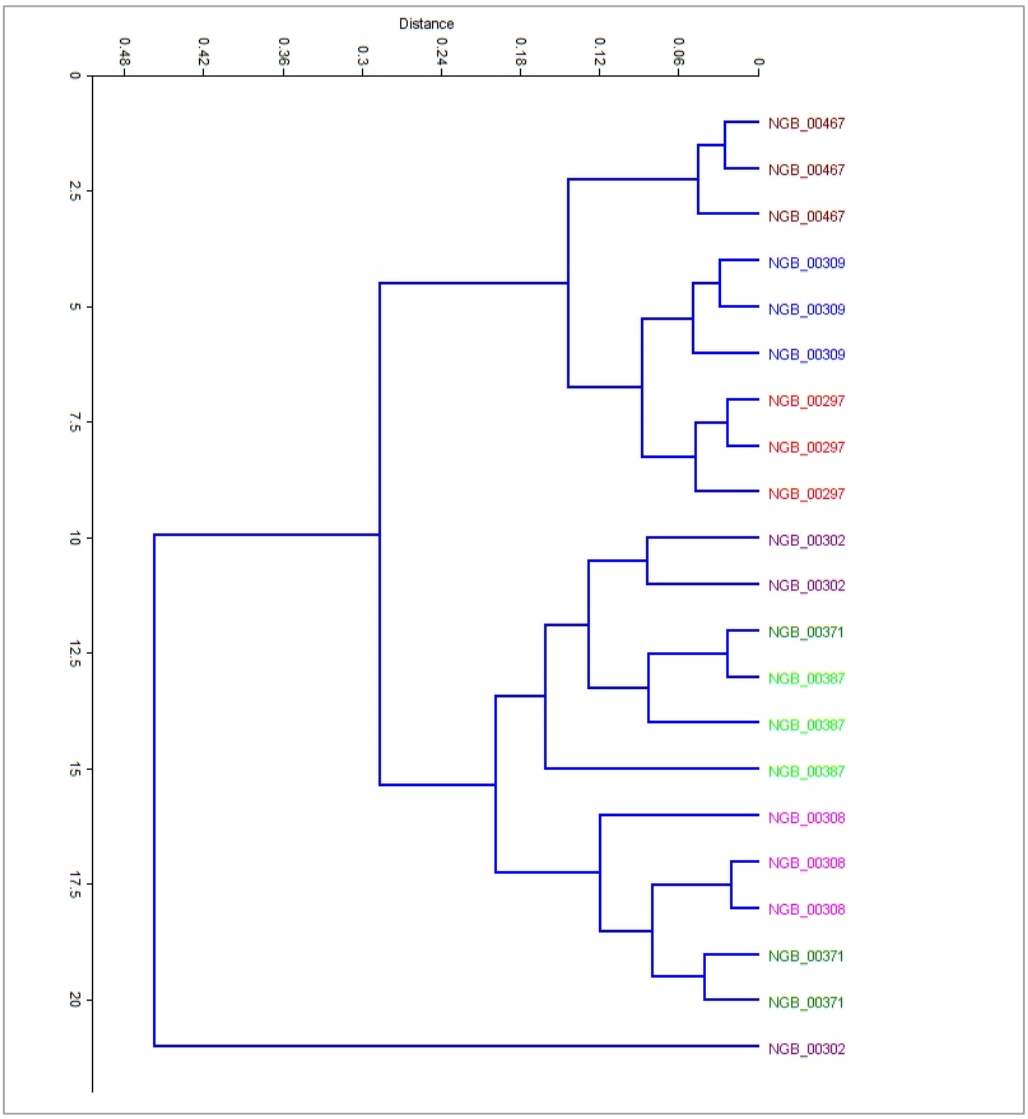

Figure 7. Gower-based UPGMA clustering analysis of the phenotypic characters of the okra accessions with a Cophenetic correlation of 0.87754.

Table 4. Pairwise correlation analysis of RBCL genotyping sequence data.

Accessions*	**00302[a]**	**00371[a]**	**00387[a]**	**00387[b]**	**00308[a]**	**00297[a]**	**00308[b]**	**00309[b]**	**00297[b]**	**00467[a]**	**00302[b]**
00302[a]	0	1.12	0.54	0.625	0.594	0.597	0.6	0.605	0.599	0.608	0.684
00371[a]	1.12	0	1.66	1.745	1.714	1.717	1.72	1.725	1.72	1.728	1.804
00387[a]	0.54	1.66	0	0.085	0.054	0.057	0.06	0.065	0.06	0.068	0.144
00387[b]	0.625	1.745	0.085	0	0.072	0.075	0.078	0.083	0.078	0.087	0.162
00308[a]	0.594	1.714	0.054	0.072	0	0.003	0.006	0.011	0.006	0.014	0.09
00297[a]	0.597	1.717	0.057	0.075	0.003	0	0.004	0.009	0.004	0.012	0.088
00308[b]	0.6	1.72	0.06	0.078	0.006	0.004	0	0.005	0.005	0.014	0.09
00309[b]	0.605	1.725	0.065	0.083	0.011	0.009	0.005	0	0.01	0.019	0.094
00297[b]	0.599	1.72	0.06	0.078	0.006	0.004	0.005	0.01	0	0.009	0.084
00467[a]	0.608	1.728	0.068	0.087	0.014	0.012	0.014	0.019	0.009	0	0.076
00302[b]	0.684	1.804	0.144	0.162	0.09	0.088	0.09	0.094	0.084	0.076	0

*All accession names start with NGB. Superscripts a and b represent sample replicates 1 and 2, respectively.

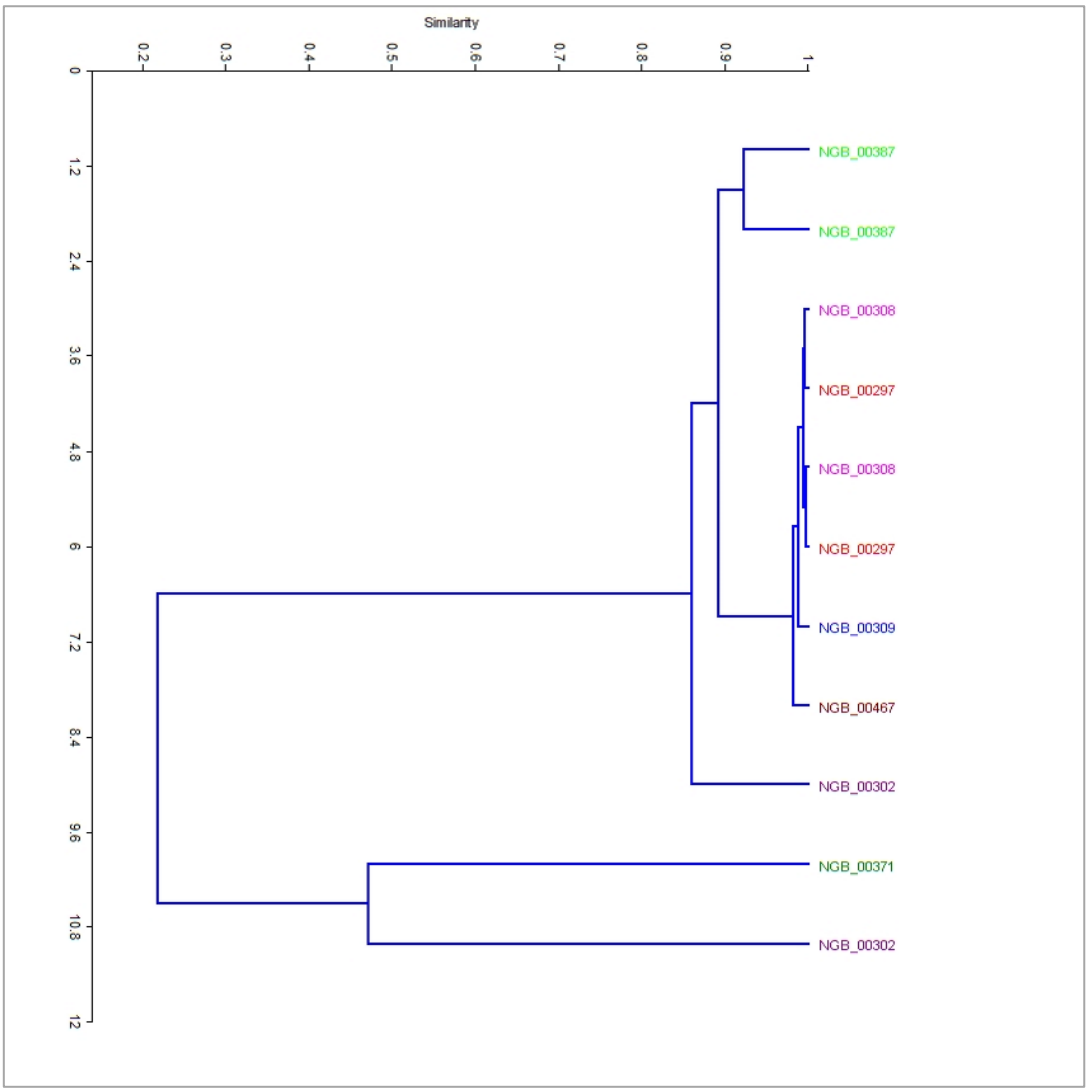

Figure 8. Bray Curtis-based UPGMA clustering dendrogram of the genetic relationship among the okra accessions with a Cophenetic correlation value of 0.981.

Discussion

The results of this study are indicative of the fact that all accessions of the Okra exhibited significant variation in morphological traits (quantitative traits), but minimal variation in qualitative traits, among the accessions investigated. This correlates with the results of (Omalsaad et al., 2014) stating that the latter traits are not useful for studying the genetic diversity of okra germplasm. Observation of significant differences among the quantitative traits is, however, an indication that genetic diversity exists among the accessions. More so, morphological characteristics are an important tool for the evaluation of plants for systematic classification and breeding. These characteristics can in some cases complement the molecular and biochemical basis of characterizing the plant germplasm although it is not enough to set them apart. From the studies, all seven accessions had an erect, and strong stem. The stem colour was green in accessions NGB 00297 and NGB 00309. Accessions NGB 00308, NGB 00467, NGB 00371 and NGB 00387 identified as okra showed red pigments on green stem; while stem pigmentation was wholly purple in accession NGB 00302. Hairiness (pubescence) was slightly less pronounced on stems of all seven accessions so there was variation in the stem pubescence of the okra accessions.

Moderate orthotropic branching was observed in accessions NGB 309, NGB 308, NGB 00297, NGB 00467, NGB 00371, NGB 00387 identified as okra, and strongly branched in NGB 302. Leaf shape was cordate in accessions NGB 00309, NGB 00308, and NGB 00371. In accessions NGB 00297 and NGB 00467 it was found to be lobed. Accessions NGB 00302 and NGB 387 identified as Okra had pinnately lobed leaf shapes. The leaf shape changed as the plant progressively germinated in all accessions. Leaf colouration was observed to be green in all accessions although red pigmentations were present on the leaf base, veins, and midribs of accessions NGB 00387 which was identified as okra, NGB 00308, NGB 00371, and NGB 00309. These findings are consistent with the reports of (Omonhinmin & Osawaru, 2005; Aladele et al., 2008; Oppong-Sekyere et al., 2011). Although a great similarity existed, there

were marked morphological variations between members of the two groups.

The quantitative attributes assessed in this study were recorded before flowering. These attributes were leaf width, length of leaf, and stem length. Hazra and Basu (2000) reported that days to bud emergence and plant height at maturity, among other morphological traits, are some of the most variable traits of okra that are necessary for selection programmes aimed at improving desirable traits. The leaves showed linear growth in length in all accessions except for accession NGB 00302. The length of leaf for accessions NGB 00371 and NGB 00387 was slightly similar. The leaf width was generally distinct for all accessions. The plant height of the accessions evaluated was also significantly different. The height of the plant can potentially affect yield as those that are taller are usually subject to wind damage in the event of heavy seasonal rains.

Results from the multivariate analysis are in concordance. A cluster phenotype of the morphological traits was observed at a 3.75 level of similarity forming two distinct clusters. The following accessions were grouped into one: NGB 00309, NGB 00308, NGB 297, and NGB 00467 with the other group having NGB 00302, NGB 00371, and the accession identified as Okra being characterized into this group (such as NGB 00387).

RBCL markers were used to determine a genetic variation within available accessions of *Abelmoschus*. The results from this work proved that RBCL markers are useful tools for genetic variation study in *Abelmoschus*. This study presented a detailed genetic distinctiveness and relationship among the accessions. A moderate level of variability was observed although accession NGB 00371 shows a much wider disparity. Variation generally, is a very important element in breeding programmes (Omonhinmin & Osawaru, 2005; Akinyele & Temikotan, 2007). The use of genetic assays in the identification of plant germplasm has become more efficient and useful for the better characterization of plant genetic resources. Hence it is clear from the result that some okra germplasm used in this study belongs to a restricted germplasm pool. However, the accessions that were found to share common similarities further show a narrow genetic base as reported by Kumar et al. (2017). High similarities among the accessions as reported by Hamon and Koechlin (1991) were expected due to their high self-pollinating properties (Hamon & Koechlin, 1991; Kumar et al., 2017). Gulsen et al. (2007) and Sharma et al. (2015) also reported a similar result establishing a 100% similarity among grapefruit cultivars. This arrayed the fact that though the accessions are independent species, there are bound genetically one to another and may share similar proximate and anti-nutrient properties which further characterizes them. The collection of this data will give the scientists, breeders, and geneticists with adequate information on the allelic similarities present in gene bank materials and to further develop more trials of these accessions. According to documented publications, this is the first report on genetic analysis using the RBCL marker in the characterization of *Abelmoschus* species in Nigeria.

Conclusion

Okra is a plant of immense importance because of the food and nutrition value as well as a source of potential raw material for diverse industrial processes. The okra accessions assessed in the current study showed variations and can be distinguished into *A. esculentus* and *A. caillei*. The diversity and distinction enumerated in the study further outline their potential and give credence to the role they play in plant breeding programmes, food security, and nutritional security among other economic utilization. To further investigate the genetic variation among plant genetic resources, there is a need to frequently evaluate and utilize okra germplasm and other crop germplasm present in gene banks by breeders, researchers, scientists, biologists, and geneticists to unravel the diverse values embedded in these genetic resources as many germplasms with these traits remain unutilized. Genetic diversity has in recent times been acknowledged as a specific area that can contribute to food and nutritional security. Thus, a better understanding will help to determine what to conserve, when, and how to conserve. The diversity indicated can further be utilized in heterosis breeding, transgressive breeding, and introgression of alien genes for specific traits. Nevertheless, the research seeks to contribute to the conservation and breeding of okra germplasm within Nigeria. It is therefore recommended that further studies be carried out to incorporate diverse characterization techniques, secondary phytochemicals, proximate nutrients, and anti-nutrient composition of the germplasm to obtain a more robust result.

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Conflict of Interest

The authors declare that there are no competing interests.

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Supplementary Information

Supplementary Table 1. Principal component summary of the morphological characteristics of the okra accessions.

PC	Eigenvalue	% variance
1	4.11067	55.124
2	1.69206	22.69
3	0.97432	13.066
4	0.343987	4.6129
5	0.277947	3.7273
6	0.055448	0.74356
7	0.002711	0.03636
8	1.47E-32	1.97E-31
9	0	0

Supplementary Table 2. Principal component summary of the morphological characteristics of the okra accessions.

Accessions	PC 1	PC 2	PC 3	PC 4	PC 5	PC 6	PC 7	PC 8	PC 9
NGB 00297	-1.2409	1.3748	0.53249	-0.76737	-0.55359	-0.10833	0.039873	4.10E-17	0
NGB 00297	-1.2063	1.3424	0.19817	0.56943	-0.44858	-0.00573	-0.04747	9.11E-17	0
NGB 00297	-1.2339	1.2835	0.34432	0.044642	-0.49674	-0.05163	-0.02713	6.67E-17	0
NGB 00309	-1.6025	-0.13603	-0.03121	0.32758	-0.55292	-0.21601	-0.06222	-8.62E-18	0
NGB 00309	-1.6974	-0.41344	-0.4172	0.77635	-0.51136	-0.24383	-0.00133	-5.04E-18	0
NGB 00309	-1.8078	-0.63985	-0.20232	-0.51153	-0.62179	-0.3831	0.10207	-6.82E-17	0
NGB 00308	-1.6799	-1.2024	0.018234	-0.76391	-0.47763	0.53448	-0.03406	-2.97E-16	0
NGB 00308	-1.5125	-1.5005	-0.42136	-0.1636	0.51227	0.24687	0.086307	-4.34E-16	0
NGB 00308	-1.6058	-1.9852	-0.44142	0.13567	0.49389	0.22675	-0.00697	-4.54E-16	0
NGB 00302	3.1883	0.81797	-0.50523	-0.12513	0.43318	-0.23098	-0.02538	4.18E-16	0
NGB 00302	1.9439	-0.36527	-0.1411	-0.8616	0.56661	-0.27898	-0.06867	9.98E-17	0
NGB 00302	3.3615	-1.8583	3.5698	0.51781	-0.22031	0.011755	0.024518	-7.15E-17	0
NGB 00467	1.8839	-0.45369	-0.71434	-0.10557	0.65811	-0.27652	0.04039	1.29E-16	0
NGB 00467	-2.7567	1.5166	1.1147	-0.12199	0.97437	0.006615	-0.01792	-5.05E-16	0
NGB 00467	-2.9027	1.002	0.66087	0.53052	1.0158	-0.03199	0.012588	-5.08E-16	0
NGB 00371	0.61083	-0.81829	-0.29272	-0.31982	-0.05836	0.093855	-0.09542	2.59E-17	0
NGB 00371	1.8175	0.26783	-0.46484	-0.22946	-0.25414	0.084282	-0.03487	3.13E-16	0
NGB 00371	0.37672	-1.591	-0.77428	0.002029	-0.0525	-0.01184	-0.02356	-5.28E-18	0
NGB 00387	1.8747	0.50583	-1.2721	1.6332	-0.05283	0.21402	0.020473	4.05E-16	0
NGB 00387	2.5215	2.9919	0.088454	-0.4375	-0.11125	0.41579	0.047518	4.71E-16	0
NGB 00387	1.6677	-0.13887	-0.84895	-0.12978	-0.24222	0.00453	0.071248	2.96E-16	0

Supplementary Table 3. Principal component summary of the morphological characteristics of the okra accessions.

Character	PC 1	PC 2	PC 3	PC 4	PC 5	PC 6	PC 7	PC 8	PC 9
GAS	0	0	0	0	0	0	0	0	1
STC	0.25356	-0.10248	0.09066	-0.03629	0.91748	-0.27104	0.020708	-1.55E-16	0
STP	7.72E-19	-2.19E-17	2.19E-17	-5.53E-18	2.79E-17	1.61E-16	1.79E-16	1	0
BRA	0.081776	-0.10983	0.36639	0.15053	-0.07926	0.0212	0.90422	-1.50E-16	0
LSH	-0.90765	0.16531	0.17886	0.039828	0.29821	0.15579	0.045547	-2.03E-16	0
LC	0.17574	-0.1575	-0.11289	-0.12321	0.21494	0.93241	0.028211	-1.94E-16	0
SL	0.15415	0.62817	-0.25198	0.7001	0.11188	0.1116	0.055102	6.84E-17	0
LL	0.09623	0.68596	-0.07446	-0.68219	0.021562	-0.01307	0.22055	2.05E-17	0
LW	0.2032	0.2453	0.86242	0.061131	-0.06193	0.14071	-0.35694	5.95E-18	0

GAS: General aspect of stem, STC: Stem colour, STP: Stem pubescence, BRA: Nature branching, LSH: Leaf shape, LC: Leaf colour, LL: Leaf length, LW: Leaf width, SL: Stem length.

Supplementary Table 4. Loadings of the NMDS axis.

Accession	Axis 1	Axis 2
NGB_00297	-0.23029	-0.00682
NGB 00297	-0.21822	0.013605
NGB 00297	-0.21551	0.011861
NGB 00309	-0.1892	-0.05276
NGB 00309	-0.19256	-0.07175
NGB 00309	-0.18955	-0.09693
NGB 00308	-0.01221	-0.17571
NGB 00308	0.063642	-0.1196
NGB 00308	0.073395	-0.14289
NGB 00302	0.21052	0.13782
NGB 00302	0.18856	0.028531
NGB 00302	0.51797	-0.10229
NGB 00467	-0.16017	0.079782
NGB 00467	-0.174	0.11852
NGB 00467	-0.16493	0.095566
NGB 00371	0.091822	-0.03085
NGB 00371	0.093821	0.057657
NGB 00371	0.10699	-0.07791
NGB 00387	0.13624	0.074822
NGB 00387	0.16732	0.22637
NGB 00387	0.096355	0.032964