CELL BIOLOGY & DEVELOPMENT
Volume 7, Number 2, December 2023 E-ISSN: 2580-4499
Pages: 89-101 DOI: 10.13057/cellbioldev/v070206

Seed phenotypic variations in cowpea, *Vigna unguiculata*, from selected open markets in Edo State, Nigeria

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Manuscript received: 19 July 2023. Revision accepted: 26 December 2023.

Abstract. *Ikhajiagbe B, Ogwu MC, Omage ZE. 2023. Seed phenotypic variations in cowpea,* Vigna unguiculata*, from selected open markets in Edo State, Nigeria. Biodiversitas 24: 89-101.* Understanding the phenotypic variation of *Vigna unguiculata* (cowpea) can facilitate sustainable utilization and support protein security goals. This study aimed to investigate the existence and level of seed phenotypic variations within and among three local cultivars of cowpea, namely Ife Brown, Ekpoma Local, and Sokoto White in Edo State, Southern Nigeria. This information will assist utilization, conservation planning, and breeding efforts. Key qualitative and quantitative characters were collected and analyzed using parametric and non-parametric tests. Results showed that there were no variations in the qualitative parameters among the seeds of cvs. Ekpoma Local and Sokoto White. However, cv. Ife Brown varied significantly, particularly in seed color. Significant variations (P>0.05) existed in the seed quantitative parameters. The seed volume was the most diverse, with a coefficient of variation of 13.15-14.14. Further, the seed volume of cv. Sokoto White was the most diverse. In terms of overall variation, the group mean sum of squares for cv. Ife Brown was 146.95, compared to 26.18 and 31.23 for cvs. Ekpoma Local and Sokoto White respectively, indicating that cv. Ife Brown was the most likely variable cultivar. There is a need for molecular characterization to ascertain the diversity observed in the cowpea seeds.

Keywords: Cowpea characterization, legume security, local cultivars, plant genetic resources, seeds diversity, *Vigna unguiculata*

INTRODUCTION

Cowpea (*Vigna unguiculata* (L.) Walp.) is a major legume crop of the global South hemisphere. The crop originated and was first domesticated in southern Africa, but is now cultivated in almost all parts of sub-Saharan Africa and parts of Asia and the Americas (Lazaridi and Bebeli 2023). It is a good source of dietary protein as well as a consistent source of income for both local and commercial farmers. As an abiotic stress tolerant and adaptable crop, cowpea is one of the oldest known human food sources with protein security roles (Herniter et al. 2020; Ifie et al. 2020; Jayawardhane et al. 2022). The seeds contain around 25% protein and 64% carbohydrate, while the young leaves, pods, and peas are high in vitamins and minerals (Simion 2018; Magashi et al. 2019; Ifie et al. 2019, 2020).

West and Central Africa is the world's largest producer of cowpea. Africa accounts for 95.9% of the total 7.4 million tons of cowpea seeds produced (Ifie et al. 2019; Ikhajiagbe et al. 2019; International Institute of Tropical Agriculture 2020). Nigeria is the world's largest producer and consumer of cowpeas. Cowpea is a staple food in Nigeria and Africa due to its hardiness, versatility and popularity (Omoigui et al. 2020).

Cowpea is the most important arable food legume in Sub-Saharan Africa (Adewale et al. 2011), and there are numerous varieties of cowpea both in and outside of Nigeria. The seed shapes, sizes, colors, texture, pigmentation, and growth patterns of cowpea varieties in Nigeria differ greatly (Iseghohi et al. 2019). Unfortunately, the ever-changing environment, volatile global economy, and intensification of low-input agricultural production have resulted in a dramatic increase in soil depletion and nutrient depletion in many Sub-Saharan regions (Magashi et al. 2019). This challenges food production and food stability, and while cowpea serves as a crop that meets global nutrient needs, this will only last so long before variation is lost due to cowpea succumbing to the consequences of climate change. Fortunately, genetic variation and abundance in cowpea may be used to create varieties that are more resistant to production constraints. As a result, in order to extend the collection, adequate awareness of genetic variation within current germplasm is needed. This allows breeding programs to pick and evolve more improved varieties quicker, not only in terms of yield but also of nutritional benefit (Magashi et al. 2019). Overall, germplasm with a greater genetic base acts as a buffer, providing resistance to climatic and other environmental changes and maintai6ning long-term food stability.

This challenges food production and food stability, especially for an essential crop like cowpea that contributes towards global nutrient needs like protein security. To this end, cowpea germplasms with a greater genetic base can act as a buffer, providing resistance to climatic and other environmental changes and maintaining long-term food stability (Nkhoma et al. 2020; Mekonnen et al. 2022).

Given this, when higher-precision diversity analysis approaches are not accessible to scientists, phenotypic assessment of genotypes for morphological classification remains a viable mechanism for identifying genetic heterogeneity within a population.

According to Oyenuga (1968), cowpea is an indigenous grain legume in Nigeria, but despite the popularity of the crop, not much is known about the morpho-genotypic variation of the crop. Nigeria is a secondary center of diversity of cowpea and the largest producer of the crop. Therefore, there is an urgent need to document and understand the morpho-genotypic variation to boost production, breeding, utilization, and conservation of the crop within key cultivation and distribution regions of the country like Edo State (Ortiz 1998; Edet and Ishii 2022). This study aimed to assess the level of variability in key seed phenotypic characteristics among three prominent *V. unguiculata* cultivars sold in Benin City, Edo State, Nigeria.

MATERIALS AND METHODS

Study area

Benin City was selected as the study area and is the state capital of Edo, which is situated in southern Nigeria. Benin City has a total area of 1,204 km^2 and is located approximately 40 km (25 miles) north of the Benin River and 320 km (200 miles) by road east of Lagos (465 miles^2). Benin City is the main hub of activity in the state, with a population of 1,782,000 as of 2021. It is also the epicentre of the Nigerian rubber industry (Osawaru et al. 2012, 2013, 2014).

Samples collection

Three cowpea (*V. unguiculata*) cultivars were purchased from three random locations within ten local markets from four local government areas in Benin City, Edo State (Figure 1; Table 1). The samples were 90 in total, 30 samples for each cultivar. The four local government areas and their respective markets include Ikpoba Okha (Santana, Ekiosa, and Oregbeni Markets), Oredo (Ugbighoko, Oba Market, and New Benin Markets), Egor (Egor and Uselu Markets) and Ovia North-East (Ekosodin and Oluku Markets). The total distance covered was 48.11 km (29.89 miles) and the GPS locations were retrieved using Garmin eTrex ® 10 handheld systems (Garmin Limited) (Figure 1; Table 1).

Morphological assessment procedure

The morphological assessments of the seeds were examined based on two categories, namely quantitative and qualitative characteristics. A total of 10 qualitative and 5 quantitative characters were scored on each of the cowpea varieties. Ten seeds of each variety from each local market were measured. Variegated testa color and moderate size describe *V. unguiculata* cv. Ekpoma Local, while cv. Sokoto White is distinguished by the pale grey testa and medium size and *V. unguiculata* cv. Ife Brown has a distinct uniform brown color except for the eyes (Figure 2).

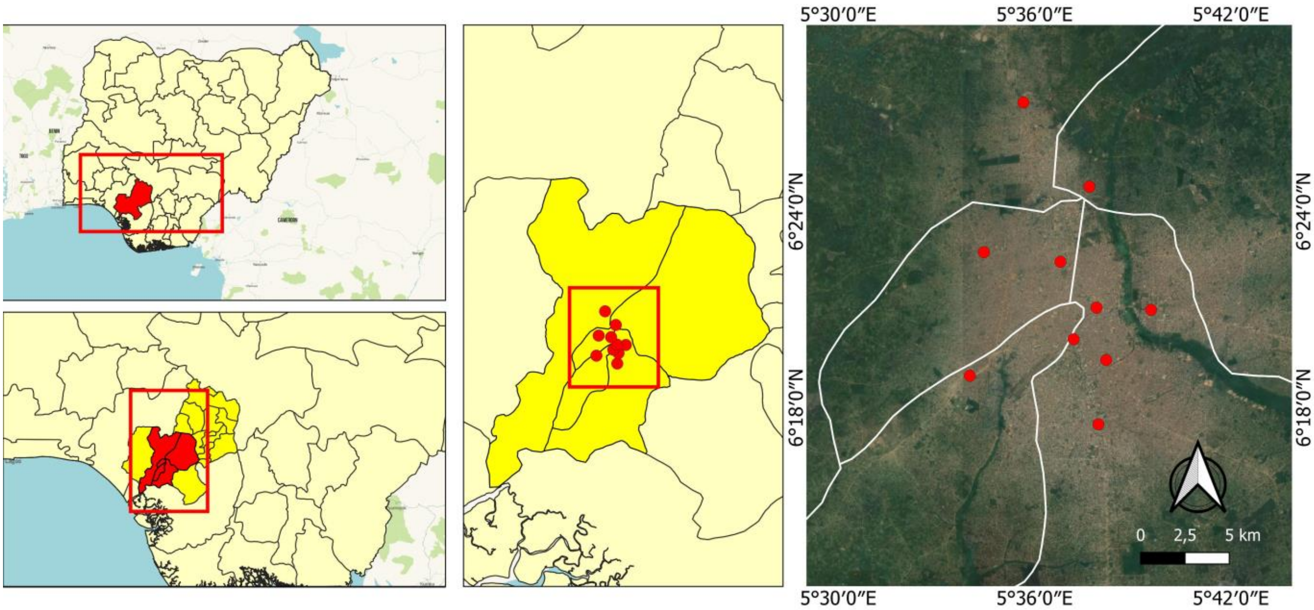

Figure 1. Map of the study area showing the source of cowpea (*Vigna unguiculata*) used in the study and distances apart

Table 1. Source of *Vigna unguiculata* seeds used in the study

Place of purchase	Local government area	Cultivar	Seed code	GPS location
Santana market	Ikpoba Okha	Ife Brown	IKCbMsL1 - IKCbMsL3	6°17'28.6" - 6°17'30.9N
		Ekpoma Local	IKCeMsL1 - IKCeMsL3	5°37'56.7" - 5°37'58.1"E
		Sokoto White	IKCsMsL1 – IKCsMsL3	
Ekiosa market	Ikpoba Okha	Ife Brown	IKCbMeL1 – IKCbMeL3	6°19'20.0" - 6°19'27.3"N
		Ekpoma Local	IKCeMeL1 – IKCeMeL3	5°38'13.0" - 5°38'11.5"E
		Sokoto White	IKCsMeL1 - IKCsMeL3	
Oregbeni market	Ikpoba Okha	Ife Brown	IKCbMoL1 – IKCbMoL3	6°20'58.7" - 6°20'59.5"N
		Ekpoma Local	IKCeMoL1 – IKCeMoL3	5°39'37.0" - 5°39'33.8"E
		Sokoto White	IKCsMoL1 - IKCsMoL3	
Ugbighoko market	Oredo	Ife Brown	ORCbMuL1 - ORCbMuL3	6°18'58.8"- 6°18'58.5"N
		Ekpoma Local	ORCeMuL1 – ORCeMuL3	5°34'03.0" - 5°34'01.4"E
		Sokoto White	ORCsMuL1 – ORCsMuL3	
Oba market	Oredo	Ife Brown	ORCbMoL1 - ORCbMoL3	6°20'03.9" - 6°20'05.3"N
		Ekpoma Local	ORCeMoL1 – ORCeMoL3	5°37'10.8" - 5°37'10.2"E
		Sokoto White	ORCsMoL1 - ORCsMoL1	
New Benin market	Oredo	Ife Brown	ORCbMnL1 - ORCbMnL3	6°21'03.0" - 6°21'04.3"N
		Ekpoma Local	ORCeMnL1 – ORCeMnL3	5°37'51.7" - 5°37'52.5"E
		Sokoto White	ORCsMnL1 – ORCsMnL3	
Ekosodin market	Ovia North-East	Ife Brown	OVCbMeL1 - OVCbMeL3	6°24'45.6" - 6°24'45.5"N
		Ekpoma Local	OVCeMeL1 – OVCeMeL3	5°37'40.8" - 5°37'40.8"E
		Sokoto White	OVCsMeL1 – OVCsMeL3	
Oluku market	Ovia North-East	Ife Brown	OVCbMoL1 - OVCbMoL3	6°27'21.1" - 6°27'20.0"N
		Ekpoma Local	OVCeMoL1 – OVCeMoL3	5°35'40.9" - 5°35'38.3"E
		Sokoto White	OVCsMoL1 – OVCsMoL3	
Uselu market	Egor	Ife Brown	EGCbMuL1 - EGCbMuL3	6°22'28.9" - 6°22'27.6"N
		Ekpoma Local	EGCeMuL1 – EGCeMuL3	5°36'50.4" - 5°36'47.6"E
		Sokoto White	EGCsMuL1 – EGCsMuL3	
Egor market	Egor	Ife Brown	EGCbMeL1 - EGCbMeL3	6°22'44.3" - 6°22'45.2"N
		Ekpoma Local	EGCeMeL1 – EGCeMeL3	5°34'28.7" - 5°34'27.1"E
		Sokoto White	EGCsMeL1 – EGCsMeL3	

Note: Local government Area: Ikpoba Okha [IK], Oredo [OR], Egor [EG], Ovia North-East [OV]; **Varieties** [C]: Ife brown [Cb], Ekpoma local [Ce], Sokoto white [Cs]; **Cultivars** [C]: Ife brown [Cb], Ekpoma local [Ce], Sokoto white [Cs]; **Markets** [M]: Santana market [Ms], Ekiosa market [Me], Oregbeni market [Mo], Ugbighoko market [Mu], Oba market [Mo], New Benin market [Mn], Ekosodin market [Me], Oluku market [Mo], Uselu market [Mu], Egor market [Me]; **Location** [L]: Location [L1], Location [L2], Location [L3]

Quantitative characteristics

Key quantitative characteristics parameters measured included seed length, seed width, seed thickness, 10-seed weight, and 20-seed volume. This was carried out using methods previously described by Osawaru et al. (2012, 2013, 2014), Chime et al. (2017), Ogwu et al. (2018), Aiwansoba et al. (2019), and Obongodot et al. (2022) by using the measurement from 10 common-sized seeds with the aid of a Uline digital venire caliper (H-7352). The linear dimensions measured were in cm and their average values were calculated and recorded. 10-seed weight was determined by weighing ten seeds of common sizes with a high-precision A & D EK-6000i- Class NTEP-approved weighing scale. The 20-seed volume was achieved by the water displacement method. Twenty seeds were dropped into a 5 mL - 2 l cylinder containing 95% ethanol and 5% water. The volume displaced was recorded as the volume of the seed.

Qualitative characteristics

The modal phenotypic and qualitative parameters assessed include eye pattern, eye color, seed shape, brilliance of seeds, splitting of testa, testa texture, color variegation, basal color, pattern of variegation, and basal color of variegated seeds as clarified in the works of Ohanmu et al. (2019a) and Ikhajiagbe et al. (2020). The testa basal color was determined using the application, Color Namer®. The qualitative characters which were determined visually were scored by nominal codes from a descriptor for cowpea by The International Board for Plant Genetic Resources (1983) (Table 2).

Data analysis

Collected data were assessed to reveal their sums of squares and least significant differences (LSD) to ascertain the source of variability among seed parameters as well as two-way analysis of variance to reveal their level of significant difference. The results were presented as a mean of 10 random determinations where necessary. The SPSS® version 21 Statistical package was used for statistical analyses.

Table 2. Qualitative characters assessed in the study and their descriptive keys

Qualitative characteristics	Descriptive key
Seed shape	1 kidney, 2 ovoid, 3 crowder, 4 globose, 5 rhomboid
Splitting of testa	0 absent, 1 present
Testa texture	1 smooth, 3 smooth to rough, 5 rough (fine reticulation); 7 rough to wrinkled
Testa color variegation	0 absent, 1 present
Testa basal color	Using the application, Color Namer® and ranged from light peach, brown, sand, light brown, tan, pale peach, beige, peach, camel, pale brown, cocoa, dull orange, butterscotch, sand brown, pinkish tan, pinkish grey, to apricot
Pattern of testa variegation	1 dense black uneven spot/dot on brown background basal color with clean eye, 2 sparse black dots on creamy brown background with a concentration around the hilum, 3 patchy light brown dots on dark brown background
Basal color of variegated seeds	0 non variegated seeds, 1 cream, 2 brown, 3 black
Eye color	0 eye absent (white, cream), 1 brown splash or grey, 2 tan brown, 3 red
Eye pattern	0 absent, 1 very small, 2 kabba group (the eye fills the narrow groove all around the hilum and the body has some form of speckling and a blue hallow is found around the hilum), 3 narrow eyes (hilum ring. Eye fills the narrow groove around the hilum and spills out of this grove in front of the hilum but for a short distance but has an indistinct front margin), 4 small eye, 5 Holsten group (i.e., the eye circles the back of the hilum in a narrow ring, widens at the sides and then extends the margin of the eye is very distinct), 6 Watson group (eye encircles the back of the hilum as a narrow ring, widens at the sides and spills over the non-micropylar end of the seed with an indistinct margin. The extra width at the sides of the hilum distinguishes this group from 3, narrow eyes).
Brilliance of the seed	1 shiny, 2 medium, 3 matt

RESULTS AND DISCUSSION

Seed phenotypic quantitative characterisation

Results of the assessments of the quantitative characteristics of *V. unguiculata* cv. Ife Brown is presented in Table 3. It was observed that no significant differences in seed length, seed width, or seed thickness were observed. Seed length ranged therefore from 1.10 to 1.39 cm respectively. The seed width, on the other hand, varied from 0.79 to 0.97 cm, while seed thickness from 0.52 to 0.64 cm respectively. However, significant differences (P>0.05) were observed in the 20-seed volume as well as the dry seed weight shown in this study. Whereas the seed sample with the lowest 20-seed volume (OVCbMeL1) was sourced at Ovia North-East local government area from Ekosodin market with a volume of 4.10 mL; compared with the 20-seed volume of 8.00 mL obtained from (EGCbMuL2) Uselu market at Egor local government area. Similarly, the lowest seed weight obtained for *V. unguiculata* cv. Ife Brown was 2.72 g from Santana market at Ikpoba Okha local government area (IKCbMsL1), and highest 4.17 g from Uselu market at Egor local government area (EGCbMuL2).

The quantitative characteristics of *V. unguiculata* cv. Ekpoma Local is presented in Table 4. The results show no significant differences in all the morphological parameters measured (P>0.05). The seed length ranged from 0.78 to 0.87 cm and 0.60 to 0.71 cm for the seed width. Seed thickness varied from 0.44 to 0.47 cm, while the 20-seed volume ranged from 2.10 to 3.20 mL. No significant changes in seed weight occurred as seed weight ranged from 1.48 to 1.74 g respectively.

The results of quantitative characteristics assessment of *V. unguiculata* cv. Sokoto White is presented in Table 5. The seed length of *V. unguiculata* cv. Sokoto White ranged from 0.74 to 0.92 cm (P>0.05). No significant changes in the seeds collected from the various sampling sites were recorded. 20-seed volume was the least (2.12 mL) at the location ORCsMoL3 compared to 3.89 ml of the 10-seed volume collected at EGCsMeL3.

The measurable mean and coefficient of variation (CV) of *V. unguiculata* seeds collected from the various markets are presented in Table 6. The results showed a mean of 1.27 cm for seed length, amounting to a CV of 4.97 for cv. Ife Brown. Compared to cv. Ekpoma Local, the mean seed length was 0.81, with a CV of 3.28; whereas, for cv. Sokoto White, the seed length was similar to cv. Ekpoma Local (0.81 cm) with a CV of 5.53. The implication of this is that the variability was more in regards to seed length in cv. Sokoto White, than cv. Ife Brown before cv. Ekpoma Local. In terms of seed dry weight, the mean value of cv. Ife Brown was 3.47 g, which eventually was the highest when compared with the seed weight of cv. Ekpoma Local (1.61 g) and cv. Sokoto White (1.64 g). In terms of CV, the results showed that the seed volume of cv. Sokoto White presented the highest amount of variation. The lowest CV was recorded in the seed thickness of the cv. Ekpoma Local (2.62) (Table 6).

Figure 2. *Vigna unguiculata* seed morphology. A. cv. Ife Brown. B. cv. Ekpoma Local. C. cv. Sokoto White. Purchased from: 1 Santana market, 2. Ekiosa market, 3. Oregbeni market, 4. Ugbighoko market, 5. Oba market, 6. New Benin market, 7. Ekosodin market, 8. Oluku market, 9. Uselu market, 10. Egor market

Table 3. Quantitative parameters of *V. unguiculata* cv. Ife Brown seeds collected at sampling sites

Seed codes	Seed length (cm)	Seed width (cm)	20-Seed volume (mL)	Seed thickness (cm)	10-Seed weight (g)
IKCbMsL1	1.25	0.82	6.00	0.52	2.72
IKCbMsL2	1.24	0.81	6.00	0.53	4.10
IKCbMsL3	1.30	0.84	6.80	0.58	3.39
IKCbMeL1	1.23	0.93	6.00	0.57	3.71
IKCbMeL2	1.24	0.84	5.80	0.57	3.12
IKCbMeL3	1.22	0.87	6.00	0.55	3.41
IKCbMoL1	1.29	0.82	6.10	0.61	3.21
IKCbMoL2	1.10	0.80	5.70	0.58	3.13
IKCbMoL3	1.31	0.79	4.90	0.62	2.90
ORCbMuL1	1.28	0.82	5.00	0.61	3.17
ORCbMuL2	1.23	0.88	5.00	0.58	3.57
ORCbMuL3	1.29	0.84	5.20	0.58	3.46
ORCbMoL1	1.21	0.86	6.00	0.61	3.25
ORCbMoL2	1.26	0.87	6.00	0.60	3.38
ORCbMoL3	1.15	0.86	5.60	0.61	3.36
ORCbMnL1	1.21	0.82	6.00	0.56	3.78
ORCbMnL2	1.16	0.84	6.00	0.58	3.75
ORCbMnL3	1.34	0.86	6.30	0.57	3.52
OVCbMeL1	1.33	0.86	4.10	0.61	3.36
OVCbMeL2	1.25	0.87	6.80	0.62	3.62
OVCbMeL3	1.37	0.97	7.00	0.61	3.49
OVCbMoL1	1.31	0.84	5.70	0.55	3.85
OVCbMoL2	1.30	0.86	5.20	0.57	3.21
OVCbMoL3	1.32	0.84	5.30	0.60	3.65
EGCbMuL1	1.29	0.83	5.00	0.54	3.83
EGCbMuL2	1.39	0.84	8.00	0.59	4.17
EGCbMuL3	1.27	0.85	5.40	0.58	3.91
EGCbMeL1	1.30	0.90	5.40	0.64	3.69
EGCbMeL2	1.31	0.91	6.30	0.54	3.44
EGCbMeL3	1.26	0.88	5.00	0.55	2.91
SD	0.06	0.04	0.75	0.03	0.34
LSD (0.05)	0.69	0.31	1.04	0.16	1.26
P-value	0.172	0.581	0.043	0.077	0.016

Note: SD: Standard Deviation, LSD: Least significant difference

Table 4. Quantitative parameters of *V. unguiculata* cv. Ekpoma Local seeds collected at sampling sites

Seed codes	Seed length (cm)	Seed width (cm)	20-Seed volume (ml)	Seed thickness (cm)	10-Seed weight (g)
IKCeMsL1	0.85	0.63	2.90	0.46	1.59
IKCeMsL2	0.87	0.64	2.40	0.46	1.59
IKCeMsL3	0.84	0.59	3.00	0.47	1.49
IKCeMeL1	0.83	0.60	3.00	0.45	1.58
IKCeMeL2	0.82	0.71	2.00	0.46	1.61
IKCeMeL3	0.85	0.61	3.00	0.46	1.51
IKCeMoL1	0.86	0.65	2.70	0.45	1.55
IKCeMoL2	0.83	0.63	2.80	0.44	1.59
IKCeMoL3	0.81	0.65	3.00	0.45	1.63
ORCeMuL1	0.79	0.62	3.15	0.45	1.69
ORCeMuL2	0.79	0.62	3.00	0.45	1.67
ORCeMuL3	0.83	0.66	3.00	0.49	1.65
ORCeMoL1	0.78	0.62	3.20	0.46	1.65
ORCeMoL2	0.78	0.70	2.50	0.44	1.62
ORCeMoL3	0.79	0.61	2.80	0.44	1.71
ORCeMnL1	0.78	0.63	3.07	0.47	1.61
ORCeMnL2	0.81	0.61	2.90	0.45	1.63
ORCeMnL3	0.78	0.58	2.50	0.44	1.74
OVCeMeL1	0.79	0.63	2.00	0.45	1.63
OVCeMeL2	0.79	0.68	2.60	0.45	1.56
OVCeMeL3	0.78	0.61	2.10	0.44	1.65
OVCeMoL1	0.80	0.61	2.80	0.44	1.60
OVCeMoL2	0.81	0.63	2.20	0.43	1.48
OVCeMoL3	0.85	0.66	3.10	0.45	1.53
EGCeMuL1	0.81	0.62	2.50	0.44	1.65
EGCeMuL2	0.84	0.64	3.00	0.46	1.74
EGCeMuL3	0.81	0.57	3.11	0.45	1.68
EGCeMeL1	0.80	0.62	2.20	0.45	1.52
EGCeMeL2	0.82	0.63	2.20	0.45	1.65
EGCeMeL3	0.83	0.63	3.00	0.46	1.59
SD	0.03	0.03	0.37	0.01	0.07
LSD (0.05)	0.21	0.21	1.09	0.11	0.61
P-value	0.305	0.749	0.665	0.532	0.129

Note: SD: Standard Deviation, LSD: Least significant difference

Table 5. Quantitative parameters of *V. unguiculata* cv. Sokoto White seeds collected at sampling sites

Seed codes	Seed length (cm)	Seed width (cm)	10 Seed volume (ml)	Seed thickness (cm)	Seed weight (g)
IKCsMsL1	0.80	0.61	3.40	0.48	1.64
IKCsMsL2	0.81	0.65	2.40	0.49	1.58
IKCsMsL3	0.89	0.70	2.43	0.46	1.42
IKCsMeL1	0.89	0.63	3.00	0.48	1.67
IKCsMeL2	0.81	0.60	2.80	0.44	1.65
IKCkMeL3	0.92	0.63	3.80	0.50	1.74
IKCsMoL1	0.79	0.63	3.10	0.47	1.57
IKCsMoL2	0.77	0.61	3.00	0.44	1.66
IKCsMoL3	0.76	0.64	2.50	0.52	1.60
ORCsMuL1	0.81	0.63	3.00	0.46	1.49
ORCsMuL2	0.85	0.62	3.00	0.46	1.57
ORCsMuL3	0.78	0.62	2.50	0.45	1.66
ORCsMoL1	0.77	0.61	2.50	0.48	1.76
ORCsMoL2	0.73	0.61	3.00	0.50	1.67
ORCsMoL3	0.78	0.55	2.12	0.40	1.59
ORCsMnL1	0.74	0.61	2.90	0.48	1.64
ORCsMnL2	0.75	0.63	3.11	0.50	1.66
ORCsMnL3	0.77	0.60	3.00	0.44	1.55
OVCsMeL1	0.86	0.65	3.00	0.48	1.63
OVCsMeL2	0.87	0.66	3.60	0.49	1.60
OVCsMeL3	0.82	0.68	3.30	0.50	1.76
OVCsMoL1	0.79	0.63	2.80	0.49	1.65
OVCsMoL2	0.91	0.61	3.00	0.43	1.62
OVCsMoL3	0.76	0.61	2.60	0.46	1.47
EGCsMuL1	0.79	0.63	3.16	0.47	1.91
EGCsMuL2	0.81	0.61	3.00	0.43	1.96
EGCsMuL3	0.77	0.65	3.20	0.48	1.84
EGCsMeL1	0.87	0.70	3.50	0.52	1.48
EGCsMeL2	0.84	0.65	3.30	0.49	1.52
EGCsMeL3	0.80	0.64	3.89	0.49	1.70
SD	0.05	0.03	0.41	0.03	0.12
LSD (0.05)	0.21	0.11	1.23	0.11	0.14
P-value	0.309	0.160	0.746	0.587	0.064

Note: SD: Standard Deviation, LSD: Least Significant Difference

Table 6. Measurable mean and coefficient of variation of *V. unguiculata* seeds collected at sampling sites

Cowpea variety	Plant quantitative parameter	Mean	SD	95% C.I.		CV
				Lower bound	Upper bound	
V. unguiculata cv. Ife Brown	Seed length (cm)	1.27	0.06	1.24	1.29	4.97
	Seed width (cm)	0.85	0.04	0.84	0.87	4.47
	Seed volume (ml)	5.79	0.76	5.5	6.07	13.15
	Seed thickness (cm)	0.58	0.03	0.57	0.59	5.14
	Seed weight (g)	3.47	0.35	3.34	3.6	9.96
V. unguiculata cv. Ekpoma Local	Seed length (cm)	0.81	0.03	0.8	0.82	3.28
	Seed width (cm)	0.62	0.02	0.62	0.63	2.75
	Seed volume (ml)	2.71	0.36	2.58	2.85	13.37
	Seed thickness (cm)	0.45	0.01	0.45	0.46	2.62
	Seed weight (g)	1.61	0.07	1.59	1.64	4.19
V. unguiculata cv. Sokoto White	Seed length (cm)	0.81	0.04	0.79	0.82	5.53
	Seed width (cm)	0.63	0.03	0.62	0.64	4.81
	Seed volume (ml)	2.93	0.41	2.78	3.09	14.14
	Seed thickness (cm)	0.47	0.03	0.46	0.48	5.9
	Seed weight (g)	1.64	0.12	1.6	1.69	7.4

Note: SD: Standard Deviation, CI: Confidence Interval, CV: Coefficient of Variation

The assessment of the sum of squares in an attempt to compare the genetic capabilities and genetic characteristics of the seeds is presented in Table 7. The results indicate that in regard to the mean sum of squares when opposed to cv. Ekpoma Local and cv. Sokoto White, whereas cv. Ife Brown has the greatest variability.

Seed phenotypic qualitative characterisation

The modal phenotypic and qualitative parameters of *V. unguiculata* cv. Ife Brown is presented in Table 8. In terms of seed shape, all seed samples selected throughout the sampling sites were 5 (rhomboid). In terms of splitting of testa, all seeds were predominantly 1 (presence of testa splitting). In terms of testa texture, all the seeds were 7 (rough to wrinkled). Except for testa basal color, which had considerable variation in coloration in the testa of sample seeds, the values for pattern of testa variegation, eye color, and brilliance of seeds amongst others were largely uniform.

The modal phenotypic and qualitative characteristics of *V. unguiculata* cv. Ekpoma Local is presented in Table 9. No changes in seed shape were observed, as the seeds were generally 5 (rhomboid). In terms of testa color variegation, all seeds obtained were generally and unanimously 1 (presence of testa color variegation). The prominent testa basal color for cv. Ekpoma Local was dark brown. All the seeds were obtained from the various markets for cv. Ekpoma Local, has the same brilliance of seeds, 2 (medium). The values for the pattern of testa variegation, eye color, testa texture and eye pattern amongst others were also largely uniform.

The modal phenotypic and qualitative characteristics of *V. unguiculata* cv. Sokoto White is presented in Table 10. The findings revealed that the shape of the seeds, the splitting of the testa, the texture of the testa, and the color variegation of the testa all followed the same pattern throughout the experiment. The seeds of *V. unguiculata* cv. Sokoto White were generally pale grey in terms of testa basal color. There were no variegations in the appearance of testa variegation pattern, basal color of variegated seeds, eye color, eye pattern, and seed brilliance. That is, there were no changes in the above five parameters regardless of the market areas from which they were purchased.

Discussion

Seed quantitative and qualitative phenotypic characterization of three *V. unguiculata* cultivars collected from open markets within Edo State, Nigeria has been completed. Except of seed volume and seed weight, there were no significant differences (P>0.05) in the quantitative parameters evaluated for *V. unguiculata* cv. Ife Brown. There were also no significant differences in all the quantitative parameters assessed in cv. Ekpoma Local and cv. Sokoto White. This observation is similar to the reports of Dorvlo et al. (2022) on *V. unguiculata* var. *Videza* from Ghana wherein variations were mainly in seed weight and sizes. According to Fatokun et al. (1992), two unlinked major gene families within cowpea's quantitative trait loci genomic regions account for the majority of variations in seed volume and weight. Due to the yield and commercial value of cowpea seed (dry-grain) size, *V. unguiculata* cv. Ife Brown would likely have more income security value than cv. Ekpoma Local and cv. Sokoto White. Interestingly, cowpea seed weight, length, and weight traits are regulated by one pleiotropic locus (Lo et al. 2019). However, this was not supported by findings in the current study as there were no similar significant differences (P>0.05 in seed length and width. In addition to seed weight and volume, the report of other workers suggests the number of days to flowering, number of productive branches, pod length and width, leaf length, and width, number of seeds per pod, and number of pods per plant as key phenotypic quantitative agronomic traits of cowpea (Menssen et al. 2017; Odeseye et al. 2018; Gerrano et al. 2022).

Vigna unguiculata cv. Ife Brown had diverse testa basal colors, while the other parameters were distributed uniformly. It was the only variety that displayed testa splitting. Due to the obvious large size of cv. Ife Brown, the splitting can be traced to inadequate sorting and handling procedures. Hence, better handling procedures should be adopted. The existence of variegation, set cv. Ekpoma Local apart. The color difference in the testa was predominantly dark brown, while the other parameters were uniformly distributed. Cultivar Sokoto White also had uniformly distributed parameters. The seed volume had the highest coefficient of variation (CV) among the three varieties studied. This implies that the seed volumes for each variety have the greatest degree of heterogeneity. Although cv. Sokoto White had the highest CV for seed volume, cv. Ife Brown had the highest number of squares, while cv. Ekpoma Local had the lowest sum of squares. The difference between cv. Ekpoma Local and cv. Sokoto White was low. *Vigna unguiculata* cv. Ife Brown had the most variation. Though cv. Ekpoma Local and cv. Sokoto White are similar in size, but they differ significantly in testa basal color, variegation presence, eye color, and pattern. The pale grey testa basal color, lack of color variegation, and greyish eye color cv. Sokoto White has all been identified as significant differences. Cultivar Ekpoma Local has a dark brown basal testa color as well as a variegated testa. Phenotypically, the three cultivars assessed in the current are not the same.

There is a high chance of variations arising within legume species, such as *Medicago truncatula*, *Lotus japonicus*, *Phaseolus vulgaris*, *Arachis hypogaea*, *Cajanus cajan*, and *Cicer arietinum* and these differences can be attributed to environmental, physiological, and genetic influences (Smykal et al. 2022; Salgotra and Stewart 2022). The environmental influences also include the agricultural production preferences of the farmer. As a consequence, our results may be affected by these factors. Cultivar Ife Brown has different testa basal colors, which may be due to the expression of many color factor genes, as seed testa color expression in cowpea is regulated by many genes. Many genes are thought to be involved in the inheritance of seed testa color in cowpea and these are together called *Color Factor* and includes *Watson*, *Holstein-1*, and *Holstein-2* in a three-locus system (Egbadzor et al. 2014; Zuluaga et al. 2021).

Table 7. Assessment of the sum of squares of measured parameters of *V. unguiculata* collected at sampling sites

Source of variation	Type III sum of squares	Df	Mean square	F	*P*-value
V. unguiculata cv. Ife Brown					
Corrected Model	587.8[a]	4	146.95	1042.6	<0.001
Intercept	857.9	1	857.87	6086.6	<0.001
Group	587.8	4	146.95	1042.6	<0.001
Error	20.4	145	0.14		
Total	1466.1	150			
Corrected Total	608.2	149			
[a.]R Squared = 0.966 (Adjusted R Squared = 0.965)					
V. unguiculata cv. Ekpoma Local					
Corrected Model	104.8[b]	4	26.18	953.8	<0.001
Intercept	231.9	1	231.91	8448.3	<0.001
Group	104.7	4	26.18	953.8	<0.001
Error	3.9	145	0.03		
Total	340.6	150			
Corrected Total	108.7	149			
[b.]R Squared = 0.963 (Adjusted R Squared = 0.962)					
V. unguiculata cv. Sokoto White					
Corrected Model	124.9[c]	4	31.22	819.9	<0.001
Intercept	252.2	1	252.18	6622.2	<0.001
Group	124.9	4	31.23	819.9	<0.001
Error	5.5	145	0.04		
Total	382.6	150			
Corrected Total	130.4	149			
[c.]R Squared = 0.958 (Adjusted R Squared = 0.956)					

Table 8. Modal phenotypic and qualitative parameters of *V. unguiculata* cv. Ife Brown seeds collected at sampling sites

Seed codes	Seed shape	Splitting of testa	Testa texture	Testa color variegation	Testa basal color	Pattern of testa variegation	Basal color of variegated seed	Eye color	Eye pattern	Brilliance of seeds
IKCbMsL1	5	1	7	0	Light peach	0	0	3	3	2
IKCbMsL2	5	1	7	0	Sand	0	0	3	3	2
IKCbMsL3	5	1	7	0	Light brown	0	0	3	3	2
IKCbMeL1	5	1	7	0	Tan	0	0	2	3	2
IKCbMeL2	5	1	7	0	Light brown	0	0	2	3	2
IKCbMeL3	5	1	7	0	Light peach	0	0	2	3	2
IKCbMoL1	5	1	7	0	Pale peach	0	0	2	3	2
IKCbMoL2	5	1	7	0	Beige	0	0	2	3	2
IKCbMoL3	5	1	7	0	Light peach	0	0	2	3	2
ORCbMuL1	5	1	7	0	Tan	0	0	2	3	2
ORCbMuL2	5	1	7	0	Peach	0	0	2	3	2
ORCbMuL3	5	1	7	0	Light peach	0	0	2	3	2
ORCbMoL1	5	1	7	0	Camel	0	0	2	3	2
ORCbMoL2	5	1	7	0	Light brown	0	0	2	3	2
ORCbMoL3	5	1	7	0	Pale brown	0	0	2	3	2
ORCbMnL1	5	1	7	0	Cocoa	0	0	2	3	2
ORCbMnL2	5	1	7	0	Pale brown	0	0	2	3	2
ORCbMnL3	5	1	7	0	Light peach	0	0	2	3	2
OVCbMeL1	5	1	7	0	Dull orange	0	0	3	3	2
OVCbMeL2	5	1	7	0	Butterscotch	0	0	3	3	2
OVCbMeL3	5	1	7	0	Light peach	0	0	3	3	2
OVCbMoL1	5	1	7	0	Sand brown	0	0	3	3	2
OVCbMoL2	5	1	7	0	Sand brown	0	0	3	3	2
OVCbMoL3	5	1	7	0	Tan	0	0	3	3	2
EGCbMuL1	5	1	7	0	Pinkish tan	0	0	1	3	2
EGCbMuL2	5	1	7	0	Pinkish grey	0	0	1	3	2
EGCbMuL3	5	1	5	0	Light brown	0	0	1	3	2
EGCbMeL1	5	1	7	0	Apricot	0	0	2	3	2
EGCbMeL2	5	1	7	0	Light peach	0	0	2	3	2
EGCbMeL3	5	1	7	0	Light peach	0	0	2	3	2

Table 9. Modal phenotypic and qualitative parameters of V. *unguiculata* cv. Ekpoma Local seeds collected at sampling sites

Seed codes	Seed shape	Splitting of testa	Testa texture	Testa color variegation	Testa basal color	Pattern of testa variegation	Basal color of variegated seed	Eye color	Eye pattern	Brilliance of seeds
IKCeMsL1	5	0	3	1	Dark brown	2	1	2	6	2
IKCeMsL2	5	0	3	1	Dark brown	2	1	2	6	2
IKCeMsL3	5	0	3	1	Dark brown	2	1	2	6	2
IKCeMeL1	5	0	3	1	Dark brown	2	1	2	6	2
IKCeMeL2	5	0	3	1	Light brown	2	1	2	6	2
IKCeMeL3	5	0	3	1	Dark brown	2	1	2	6	2
IKCeMoL1	5	0	3	1	Dark brown	2	1	2	6	2
IKCeMoL2	5	0	3	1	Dark brown	2	1	2	6	2
IKCeMoL3	5	0	3	1	Dark brown	2	1	2	6	2
ORCeMuL1	5	0	3	1	Dark brown	2	1	2	6	2
ORCeMuL2	5	0	3	1	Light brown	2	1	2	6	2
ORCeMuL3	5	0	3	1	Light brown	2	1	2	6	2
ORCeMoL1	5	0	3	1	Dark brown	2	1	2	6	2
ORCeMoL2	5	0	3	1	Dark brown	2	1	2	6	2
ORCeMoL3	5	0	3	1	Dark brown	2	1	2	6	2
ORCeMnL1	5	0	3	1	Dark brown	2	1	2	6	2
ORCeMnL2	5	0	3	1	Dark brown	2	1	2	6	2
ORCeMnL3	5	0	3	1	Light brown	2	1	2	6	2
OVCeMeL1	5	0	3	1	Dark brown	2	1	2	6	2
OVCeMeL2	5	0	3	1	Dark brown	2	1	2	6	2
OVCeMeL3	5	0	3	1	Dark brown	2	1	2	6	2
OVCeMoL1	5	0	3	1	Dark brown	2	1	2	6	2
OVCeMoL2	5	0	3	1	Dark brown	2	1	2	6	2
OVCeMoL3	5	0	3	1	Dark brown	2	1	2	6	2
EGCeMuL1	5	0	3	1	Light brown	2	1	2	6	2
EGCeMuL2	5	0	3	1	Dark brown	2	1	2	6	2
EGCeMuL3	5	0	3	1	Dark brown	2	1	2	6	2
EGCeMeL1	5	0	3	1	Dark brown	2	1	2	6	2
EGCeMeL2	5	0	3	1	Dark brown	2	1	2	6	2
EGCeMeL3	5	0	3	1	Dark brown	2	1	2	6	2

According to Tiryaki et al. (2016), the testa color trait is polygenic and influenced by multiple genes in a variety of plant species, including legumes such as cowpea, common bean (*Phaseolus vulgaris*), and soybean (*Glycine max*). This expression of multiple genes results in varying levels of several color pigments in the seed testa, which explains the observed color changes in seed testa (Mavi 2010). Environmental factors like temperature and light intensity may also affect the production of these pigments II (Ohanmu et al. 2019b). According to Bhatt et al. (2016), seed color has also been stated to play a role in seed dormancy and germination in leguminous plants. As a result, further research should be conducted to examine the relationship between seed testa color and dormancy as well as germination. Furthermore, seed size and seed coat color have been used to establish a simple method of improving seed quality for many crop species, including common bean (*P. vulgaris*), cowpea, rapeseed (*Brassica napus*), flax (*Linum usitatissimum*), and *Arabidopsis thaliana* (Tiryaki et al. 2016). In contemporary agricultural systems, where uniformity is preferred, seed differences may cause uncertainty (Mitchell et al. 2016). More so if these differences like seed color, weight, shape, and volume have negative impacts on crop yield like reducing yield numbers and amounts. These seed differences may be unfavourable to both the seller and buyer, particularly when they are undesirable traits to the end users and consumers.

Over 115 common bean germplasm resources were assessed using key morphological characters and it was discovered that the population was highly diverse (Long et al. 2020). A study of the phenotypic diversity of two chickpeas (*Cicer arietinum*) collections was conducted in Ethiopia where data were obtained from three independent places in one region, and the results indicated significant differences in phenotypic and agronomic performance variability between the two collections (Admas et al. 2021). Another research looked at the variance in seed morphologies of 160 *Cucurbita maxima* populations obtained from different parts of Turkey. Sizeable differences in seed shape, color, size, and weight were ascertained (Balkaya et al. 2009). In the study of 56 Japanese native cultivars of common buckwheat (*Fagopyrum esculentum*), a considerable number of variances in seed shape characteristics and husk colors were also detected (Tetsuka and Uchino 2005).

Table 10. Modal phenotypic and qualitative parameters of *V. unguiculata* cv. Sokoto White seeds collected at sampling sites

Seed codes	Seed shape	Splitting of testa	Testa texture	Testa color variegation	Testa basal color	Pattern of testa variegation	Basal color of variegated seed	Eye color	Eye pattern	Brilliance of seeds
IKCsMsL1	5	0	5	0	Pale grey	0	0	1	2	2
IKCsMsL2	5	0	5	0	Pale grey	0	0	1	2	2
IKCsMsL3	5	0	5	0	Pale grey	0	0	1	2	2
IKCsMeL1	5	0	5	0	Pale grey	0	0	1	2	2
IKCsMeL2	5	0	5	0	Pale grey	0	0	1	2	2
IKCkMeL3	5	0	5	0	Pale grey	0	0	1	2	2
IKCsMoL1	5	0	5	0	Pale grey	0	0	1	2	2
IKCsMoL2	5	0	5	0	Pale grey	0	0	1	2	2
IKCsMoL3	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMuL1	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMuL2	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMuL3	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMoL1	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMoL2	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMoL3	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMnL1	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMnL2	5	0	5	0	Pale grey	0	0	1	2	2
ORCsMnL3	5	0	5	0	Pale grey	0	0	1	2	2
OVCsMeL1	5	0	5	0	Pale grey	0	0	1	2	2
OVCsMeL2	5	0	5	0	Pale grey	0	0	1	2	2
OVCsMeL3	5	0	5	0	Pale grey	0	0	1	2	2
OVCsMoL1	5	0	5	0	Pale grey	0	0	1	2	2
OVCsMoL2	5	0	5	0	Pale grey	0	0	1	2	2
OVCsMoL3	5	0	5	0	Pale grey	0	0	1	2	2
EGCsMuL1	5	0	5	0	Pale grey	0	0	1	2	2
EGCsMuL2	5	0	5	0	Pale grey	0	0	1	2	2
EGCsMuL3	5	0	5	0	Pale grey	0	0	5	6	2
EGCsMeL1	5	0	5	0	Pale grey	0	0	1	2	2
EGCsMeL2	5	0	5	0	Pale grey	0	0	1	2	2
EGCsMeL3	5	0	5	0	Pale grey	0	0	1	2	2

Benin City, Edo State (Nigeria) where the majority of the samples used in the study were sourced is a central hub where people travelling from and to diverse parts of the country go through, it is imperative to note that diverse *V. unguiculata* cultivars from different part of the country may have a legacy effect and influence local cultivars like *V. unguiculata* cvs. Ife Brown, and Ekpoma Local. Concerning the current lack of data on cowpea diversity, utilization, breeding, and conservation in Nigeria, a diverse array of criteria is needed to resolve this. Addressing the issue will enable an understanding of the source and extent of variations as well as a correlation of environmental variations within cowpea cultivars and varieties (Ifie et al. 2019; Iseghohi et al. 2019).

In conclusion, *V. unguiculata* cv. Ife Brown sold in markets within Edo state have the greatest phenotypic variations among all the cultivars accessed in this study, even though, it does not originate from and is not native to the state. The observed phenotypic variation in *V. unguiculata* cv. Ife Brown is likely due to a combination of genetic and environmental factors from the western part of Nigeria where it is native. Although, *V. unguiculata* cv. Ekpoma Local is considered native to Edo State, and has many similarities with *V. unguiculata* cv. Sokoto White, it may not originally be from the State. Also, the similarities between both cultivars may be linked to the nearly similar environmental conditions prevalent in Ekpoma and Sokoto. Generally, the findings from this study support the possibility of variations existing within the cowpea seeds available in Edo State, Southern Nigeria. This finding will enable future workers to effectively compare *V.* unguiculata through both morphological and molecular investigations.

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