



Varietal differences in seed germination and seedling vigour characteristics in local and improved cowpea genotypes

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Abstract

An experiment was conducted using twenty-two (accession, local and improved) cowpea genotypes evaluated for their laboratory seed quality attributes. The laboratory experiment was set-up in a complete randomized design with three replicates between February and March, 2023 at the Institute of Agricultural Research and Training, Obafemi Awolowo University, Moor Plantation, Ibadan, Oyo State. Data collected on seed quality attributes in the laboratory were subjected to analysis of variance. Treatment means were separated using Duncan Multiple Range Test at 5 % level of probability, correlation, and principal component analysis. Germination percentage ranged from 94.0 % for NGB07614 to 31.33 % for NGB07593. Germination percentage also had highly significant negative correlation with germination index, abnormal seedling, dead seed and seedling dry weight. Germination percentage also had positive and significant correlation with root length and seedling vigour index. Principal component analysis revealed that the seed quality attributes such as germination percentage, germination index, seedling vigour index, shoot length, root length and seedling dry weight contribute significantly to the variation within the 22 genotypes of cowpea evaluated. The cluster analysis for seed quality attributes included in this study placed cowpea genotypes into four clusters with sub clusters for each, except cluster four with only three genotypes of one accession, one local and one improved, respectively. The mean performance of laboratory seed quality attributes revealed that NGB07614, Abewere, 150-Ex and Modupe were outstanding in some of seed quality attributes. This shows that selection for superior seed quality traits is possible among these cowpea accessions.

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Introduction

Cowpea (*Vigna unguiculata* L. Walp) is a principal food and cash crop legume cultivated in the semi-arid tropics covering Africa, Asia and Central America. The crop has great socio-economic, cultural and nutritional importance (Dariba, 2012). Relatively poor communities in developing countries particularly in the tropical Africa depend on cowpea as the main food legume in supplying protein in their diets (Timko and Singh, 2008). It is mostly produced and consumed in Asia and tropical Africa (Alpha *et al.*, 2016). Cowpea is grown on about 11.32 million hectare Worldwide, with an annual grain production of about 5.72 million tons (FAO), 2014). Africa is responsible for 94 % of this total product. Nigeria is the largest cowpea producers in the world and account for over 2.5 million tons grain production from an estimated 4.9 million hectare (FAO, 2014).

Seeds are one of the greatest assets of agricultural activity in the world. Their physiological quality defines both the establishment of the crop and its productivity (Begatali *et al.*, 2019; Strucker *et al.*, 2019). High-quality seeds are more capable of generating uniform seedlings that will give rise to individuals with ample productive potential and efficient use of the resources of the environment (Caverzan *et al.*, 2018). On the other hand, seeds with reduced quality will compromise the emergence of seedlings, and result in an uneven establishment in the field, which in turn will result in low yield (Abati *et al.*, 2017; Ebone *et al.*, 2020) Cowpea production is wholly dependent on seed as propagation material. The quality of cowpea seed is therefore, an essential determinant of final quantity and quality of cowpea leaves and grain (Abukutsa-Onyango, 2011). Yield of cowpea in sub-Saharan Africa has been very low with just about 22% of the potential yield obtained on farmers' fields. The poor quality of seeds used by farmers has been identified as a limiting factor contributing to low yields in cowpea (Odindo, 2007 (Unpublished). Availability of quality seeds of improved, high yielding varieties to local farmers is still a bottleneck for increasing the grain yield of cowpea. Seed is an important input for agricultural production. Improving the availability of high-quality

seeds of improved varieties is important to boost agricultural productivity, leading to higher farmers' income, reduced poverty and improved food security (Abdoulaye *et al.*, 2009).

Seed quality includes genetic quality, physical quality, physiological quality and seed health (Louwaars, 2007 (Unpublished)). Physiological quality refers to the seeds' ability to germinate, emerge from the soil and form a vigorous seedling, especially under the stressful conditions prevailing in the fields of most smallholder farmers. High physiological quality is essential to create an optimal and uniform plant density with minimum seeding rates.

Physiological quality is influenced by environmental conditions during crop growth, harvesting and storage conditions (Ghassemi Golezani and Mazloomi-Oskooyi, 2008). Germination capacity and physiological vigour are intrinsic properties of the seed that affects its quality. It is important to classify the range of variability among accessions to facilitate the maintenance and further acquisition of germplasm resources. The objectives of the study were, therefore, to determine varietal differences in seed germination and seedling vigour quality characteristics of local and improved cowpea genotypes and to determine association among seed quality attributes of local and improved cowpea

Materials and methods

Seeds of eleven (11) cowpea accessions, five (5) local and six (6) improved cowpea varieties were used for the experiment. The seeds were sourced from National Centre for Genetic Resource and Biotechnology (NACGRAB), local farmers in Oyo North and Institute of Agricultural Research and Training, Moor Plantation, Ibadan. The seed were planted between August and December, 2022 in Ibadan research field of the Institute. The plants were harvested processed and stored in paper envelop and placed under room temperature (28°C and RH 65%) in December of the same year. The laboratory analysis was carried out in the Institute seed quality control unit between February and March, 2023

Seed Quality Assessment

The quality of seeds of the 22 selected cowpea genotypes were assessed with the following tests in the seed testing laboratory of the Institute of Agricultural Research and Training, Moor Plantation, Ibadan. Standard germination test was carried out in three replications of 100 seeds per replicate.

Plastic germination bowls were filled with moistened sharp sand and seeds were evenly spaced on the sand. Thereafter the seeds were thinly covered with moistened sand and lightly pressed for a good seed-substratum contact. The bowls were covered with nylon sheets to prevent evaporation. Germination counts were taken daily from the 3rd to 8th day after planting. On the 9th day, seedling analysis was carried out and the numbers of normal and abnormal seedlings were recorded. Germination was interpreted as the percentage of seeds producing normal seedlings (International Seed Testing Association, 2018).

$$\text{Germination percentage (GPCT)} = \frac{\text{Number of normal seedlings emerged}}{\text{Total number of seedsplanted}} \times 100$$

From the germination data above, germination index (GI) was calculated for each replicate according to Ajayi and Fakorede (2000) as follows.

$$\text{Germination index (GI)} = \frac{\sum(N_x) \text{ DAP}}{\text{Total number of seedlings that emerged on the final day}}$$

Where N_x is the number of seedlings that emerged on day x after planting.

DAP is the days after planting.

100 Seed Weight

A 100- seed weight in 3 replicates from each genotype was determined and expressed in gramme

Evaluation of seedling traits

One out of every 10 normal seedlings, that is, 10% of the total number of normal seedlings, in each replicate, at the final germination count, was used to obtain data on the following seedling vigour parameters.

Seedling shoot length (SLT)

The length from the shoot level to the shoot tip of seedling in 10 randomly selected seedlings and expressed in cm.

Root Length (RLT)

The length from shoot level to the tip of the plant root of seedling in 10 randomly selected seedlings and expressed in cm.

Seedling Dry Weight (SDWT)

10 randomly selected seedlings oven dried at 108° C until constant weight and averaged.

Seedling Vigour Index (SVI)

The seedling vigour levels of each genotype was calculated by multiplying percent seed germination by average of seedling root and shoot length of each variety after 7 days of germination and divided by 100.

$$\text{Seedling Vigour Index (SVI)} = \frac{\% \text{ germination} \times (\text{root length} + \text{shoot length})}{100}$$

Data analysis

Data collected in the laboratory were subjected to analysis of variance. Treatment means were separated using Duncan Multiple Range Test (DMRT) at 5 % level of probability. Correlation coefficients among all traits evaluated were computed. Principal Component analysis (PCA) is a tool used in exploratory data analysis and for making predictive models, achieved by Eigen value (Jolliff, 2002). It is a technology that identifies plant characters which contribute most to the variation within a group of entries. PCA is useful for finding new, more informative and uncorrelated feature. PCA was used to determine characteristics accounting for major variations among the entries. Cluster analysis was done on all the 22 genotypes based on Euclidean distance and similarity matrix to generate dendrogram (Mohammadi and Prassnna (2003).

Results

The result shows that genotype effect was significant for all the characters evaluated with the exception of number of abnormal seedlings. However, the

replication effect was not significant on any of the quality attributes evaluated in the laboratory (Table 1). Hundred seed weight ranged from 10.03g (150-EX) to 30.81g (NGB05595). NGB07593, NGB05529, NGB02183, NGB07632, NGB05595 had the highest weight of between 26.77g and 30.81g, followed by Oloyin, Kawoleri, NGB07592, NGB07614 with seed weight of between 20.27g and 22.77g while 150-EX recorded the lowest seed weight of 10.03g (Table 2).

Germination percentage ranged from 94.0 % for NGB07614 to 31.33 % for NGB07593. Other genotypes that had germination percentage of 80 % and above were Abewere, 150-EX, NGB04654, BBT-Brown, NGB07632, NGB07592, Modupe and Olomoyoyo with values of 91.3, 90.0, 88.67, 88.0, 87.33, 86.0, 84.67 and 82.05, respectively.

For germination index, genotypes Abewere, BBT-Brown, NGB04654, Modupe, Olomoyoyo and NGB07614 recorded germination index of less than 3 days after planting. Others recorded germination index of between 3, 02 DAP for 150-EX and 9.29 (Kawoleri). Genotypes NGB07589, NGB07593, NGB02183 and Kawoleri recorded high abnormal seedlings count with 14.0, 10.0, 9.33, 8.67 and 8.00%, respectively.

The least abnormal seedling was recorded in 150-EX (0.67) followed by genotypes NGB07614 (1.33), Oloyin (1.33) and NGB04654 (1.33). Genotypes Oloyin, NGB07593, Kawoleri, NGB05595, NGB02183 and BBT-White had greater percentages of dead seeds with values of 61.33, 59.67, 53.33, 50.33 and 40.67, respectively. Seedling dry weight ranged from 0.087 (NGB04654) and 0.29 (NGB07589).

Table 2 also showed that Modupe recorded highest (15.47cm) followed by NGB04688 (14.80cm) while NGB02183 recorded the lowest value of 7.20cm. Seedling shoot length of Oloyin, BBT-White and 150-EX were the highest with 26.47cm, 26.30 and 26.20cm, respectively. These were followed by NGB07593 (25.53cm), Modupe (25.44cm) and Ife-Brown (24.37cm). The lowest shoot length was

recorded by NGB02183 with the value of 13.1cm. For seedling vigour index, 150-EX had the highest index of 35.53 followed by Modupe (34.69) while NGB02183 showed the least vigour index of 8.20.

Table 1. Mean square values of the laboratory seed quality attributes evaluated in 22 cowpea genotypes.

Attributes	Replication	Genotype Mean Square	Error
100 Seed Weight	2.31	130.56**	0.85
Germination	111.31	1337.31**	87.53
Germination Index	3.33	12.05**	2.41
Abnormal Seedling	3.63	35.12	14.68
Dead Seed	91.01	1090.80**	86.44
Seedling dry weight	0.003	0.009*	0.003
Root Length	6.78	13.75*	5.10
Shoot Length	3.26	34.29**	8.10
Seedling Vigour Index	14.11	191.82**	18.50
Df	2	21	42

*,** Significant at $p < 0.05, 0.01$, respectively

Table 3 shows the correlation among seed quality attributes evaluated in cowpea genotypes. The result shows that 100 seed weight had positive and highly significant association with germination index ($r=0.455^{**}$), abnormal seedling ($r=0.46^{**}$), dead seed ($r=0.387^{**}$), seedling dry weight ($r=0.43^{**}$) but negative and highly significant correlation with germination percentage ($r=-0.457^{**}$), root length ($r=-0.259^*$), shoot length ($r=-0.463^{**}$) and seedling vigour index ($r=-0.584^{**}$).

Germination percentage had highly significant negative correlation with germination index ($r=-0.843^{**}$), abnormal seedling ($r=-0.489^{**}$), dead seed ($r=-0.978^{**}$) and seedling dry weight ($r=-0.33^{**}$). Germination percentage also had positive and significant correlation with root length ($r=0.374^*$) and seedling vigour index ($r=0.897^{**}$).

Germination index had positive and highly significant association with abnormal seedling ($r=0.432^{**}$), dead seed ($r=0.823^{**}$), root length ($r=0.375$) and seedling vigour index ($r=0.897^{**}$). Abnormal seedling had negative and significant association with seedling vigour index ($r=-0.859^{**}$) while root length had highly significant and positive association with seedling vigour index ($r=0.631^{**}$)

Table 2. Mean performance of laboratory seed quality attributes in 22 cowpea genotypes.

Genotypes	100SWT	G %	GI	ABN	DS	SDWT	RLT	SHLT	SVI
NGBo5595	30.81a	39.33jkl	8.2ab	7.33abcde	53.33ab	0.20ab	12.20abcdef	17.27ef	11.63j
NGBo7632	30.56a	87.33abcd	3.31de	4.67bcdefg	8.00ijk	0.20ab	12.47abcd	17.63ef	25.41cdefg
NGBo7589	28.41b	50.67ghij	5.49cd	14.0a	35.33cdef	0.29a	8.53fgh	18.60de	14.63hij
NGBo2183	28.40b	41.00ijkl	6.53bc	8.67abcd	50.33abc	0.18bc	7.23h	13.10f	8.20j
NGBo5529	27.37bc	62.67fgh	4.06cde	10.00ab	27.33defgh	0.18bc	10.44cdefgh	23.40abc	21.16efgh
NGBo7593	26.77c	31.33l	8.32a	9.33abc	59.33a	0.18bc	7.40gh	25.53ab	10.21j
NGBo7614	22.77d	94.0a	2.94de	1.33fg	4.67k	0.13bcd	10.67cdefgh	21.27bcde	30.06abc
NGBo7592	21.80d	86.00abcd	3.60de	3.33cdefg	10.66ijk	0.11bcd	12.40abcde	18.60cd	26.73cdef
Kawoleri	21.53de	35.33kl	9.29a	8.00bcdefg	56.67a	0.20ab	11.20bcdef	22.57abcd	11.99j
Oloyin	20.27ef	37.33kl	6.33bc	1.33fg	61.33a	0.18bc	11.17bcdef	26.47a	14.04ij
Sadu	18.92fg	72.67def	3.29de	4.67bcdefg	22.67efgh	0.13bcd	8.70efgh	22.80abcd	22.90defg
NGBo4688	18.49g	73.33cdef	4.22cde	6.00bcdefg	20.67fghij	0.15bcd	14.80ab	23.77abc	28.29bcde
Gombe	16.66h	75.33abcde	3.33de	6.00bcdefg	18.67ghijk	0.11bcd	10.70cdefgh	24.03abc	26.11cdefg
Ife-BPC	15.02i	69.33efg	4.13cde	4.00bcdefg	30.00defg	0.28a	13.97abc	23.33abc	25.80cdefg
BBT-White	14.99i	54.67ghij	4.59cde	4.67bcdefg	40.67bcd	0.15bcd	9.13defgh	26.30a	19.45ghi
Abewere	14.45i	91.3a	2.68e	2.00efg	6.67jk	0.13bcd	10.83cdefgh	19.80cde	28.08bcde
Ife Brown	14.01ij	56.00ghi	4.26cde	6.67bcdefg	37.33cde	0.13bcd	11.43bcdef	24.37abc	20.32ghi
NGBo4654	12.87j	88.67abc	2.81e	1.33fg	10.00ijk	0.08d	11.03cdefg	21.93abcde	29.29abcd
Olomoyoyo	12.80j	82.00abcde	2.90e	2.67defg	15.33ghijk	0.11bcd	11.80abcdef	21.43bcde	27.05cdef
BBT-Brown	12.79j	88.0abcd	2.75e	2.67defg	9.33ijk	0.13bcd	11.33bcdef	22.60abcd	29.86abcd
Modupe	12.49j	84.67abcde	2.86e	2.67defg	12.66hijk	0.09cd	15.47a	25.44ab	34.69ab
150-Ex	10.03k	90.00ab	3.02de	0.67g	9.33ijk	0.11bcd	13.17abc	26.20a	35.53a
LSD	1.52	15.37	2.56	6.27	15.32	0.10	3.72	4.69	7.09

Table 3. Correlation coefficient among laboratory seed quality attributes evaluated in 22 cowpea genotypes (N=69).

	100SWT	G %	GI	ABN	DS	SDW	RLT	SHLT	SVI
100SWT		-0.457**	0.455**	0.460**	0.387**	0.433**	-0.259*	-0.463**	-0.584**
G%			-0.843**	-0.489**	-0.978**	-0.332*	0.374*	-0.059	0.897**
GI				0.432*	0.823**	0.322	0.375*	-0.059	0.897**
ABN					0.304	0.292	-0.251	-0.161	-0.496**
DS						0.317	-0.337	0.108	-0.859**
SDW							0.052	-0.051	-0.289
RLT								0.244	0.631**
SHLT									0.301
SVI									

100SWT: 100 Seed Weight, G%: Germination Percent, GI: Germination Index, ABN: Abnormal Seedlings, DS: Dead Seed, SDW: Seedling Dry Weight, RLT: Root Length, SHLT: Shoot Length, SVI: Seedling Vigour Index.

Principal component analysis shows the character that contributed to variation within the genotypes. Table 4 presents the principal component for seed quality attributes in twenty-two cowpea genotypes. From the result, three principal component axes were identified with cumulative variance of 53, 68 and 80 % in PC 1, PC2 and PC3, respectively. In PC1, germination percentage (0.41) followed by germination index and dead seed with 0.40 and 0.39, respectively contributed majorly to the variation within the genotypes with Eigen value of 5.26 and variance percentage of 53. In PC 2, shoot length (-0.64) and 100 seed weight (0.43) contributed largely to the variability. However, for PC3, seedling dry weight (-0.61) and dead seed (-0.69) contributed the largest variability in the genotypes.

Table 4. Principal component analysis based on correlation coefficient matrix for the seed quality attributes evaluated in cowpea.

Principal Component	PC1	PC2	PC3
Eigen values	5.26	1.54	2.26
% Variance	53	15	11
% Cumulative Percentage	53	68	80
100SWT	-0.28	0.43	-0.27
Germination	-0.41	0.19	-0.05
Germination Index	-0.40	-0.19	-0.05
Abnormal Seedlings	-0.25	0.22	-0.18
Dead Seed	-0.39	-0.26	0.08
Seedling Dry Weight	-0.18	0.08	-0.69
Root Length	0.20	-0.27	-0.61
Shoot Length	0.07	-0.64	-0.06
Seedling Vigour Index	0.41	-0.11	-0.21

Based on the dendrogram drawn from single linkage cluster analysis Table 5 and Figure 1, the cowpea genotypes at 0.0 level of similarity were all distinct from each other. But at 34.0 level of similarity, the

genotypes formed four major clusters. Cluster 1 contain three (3) genotypes which are BBT-White, Ife-BPC, and Ife-Brown, the second cluster contain three genotypes NGB05595, NGB07589, and NGB02183. Cluster III contain thirteen genotypes NGB04654, NGB04688, 150-EX, NGB05529, Abewere, BBT-Brown, NGB07592, Gombe, NGB07614, NGB07632, Olomoyoyo, Modupe, and Sadu while the fourth cluster comprises three genotypes NGB07593, Kawoleri, and Oloyin. Genotypes that are clustered together share similar attributes and belong to the same base population.

Table 5. Cluster distribution of twenty-two cowpea genotypes based on UPGMA Dendrogram.

Cluster	Genotypes	Number of Genotypes
I	BBT-White, Ife-BPC, and Ife-Brown	3
II	NGB05595, NGB07589, and NGB02183	3
III	NGB04654, NGB04688, 150-EX, NGB05529, Abewere, BBT-Brown, NGB07592, Gombe, NGB07614, NGB07632, Olomoyoyo, Modupe, and Sadu	13
IV	NGB07593, Kawoleri, and Oloyin	3

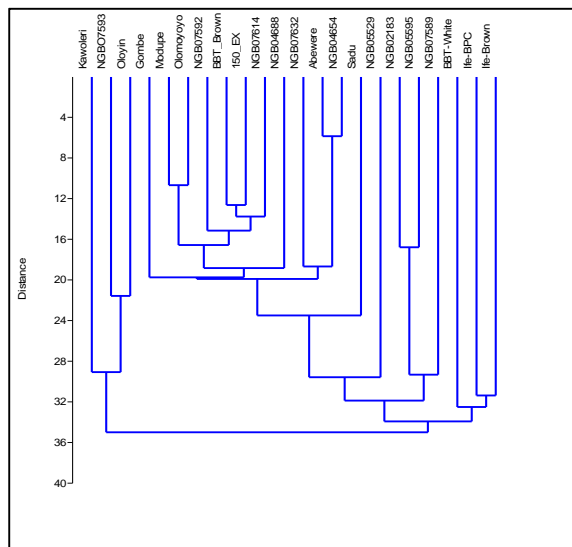


Fig. 1. Dendrogram showing clustering pattern of twenty-two (22) cowpea genotypes based on seed quality attributes.

Discussion

Variability among accession is fundamental to the maintenance and further acquisition of germplasm

resources even as accessions from diverse origins are needed as parent stocks for the development of improved varieties (Zenabou *et al.*, 2014). This necessitates the evaluation of varietal differences seed germination and seedling vigour differences in 22 accessions of cowpea. From the result of this study, the genotypes of cowpea evaluated differed significantly for all the seed physiological quality attributes. The differences may be attributed to diverse genetic background of the cowpea accessions studied. This implies that the existence of variability among the genotype for these traits will give opportunity for selecting genotypes of cowpea with superior seed quality attribute. Significant differences in seed quality attributes among genotypes have been reported by different authors in different crop species (Abdul- Rafiu, 2014 (Unpublished) in pepper and Alegiledoye, 2016 (Unpublished) in African Yam Beans). Significant differences in 100 seed weights are similar to the findings of Kabambe *et al.* (2014) who found that IITA-improved varieties were superior in term of seed weight. This also agreed with the findings of Ehler and Hall (1997) who stated that in most of the cowpea lines, the seed size varies, some varieties weigh less than 10g per 100 seed and some weigh approximately 30g. The mean performance of laboratory seed quality attributes revealed that NGB07614, Abewere, 150-Ex and Modupe were outstanding in term of seed quality attributes. This shows that selection for superior seed quality traits is possible among these cowpea accessions. This finding supports the observation of Ajala (2003) in pea. Relationship among the laboratory seed quality attributes result shows that 100SWT had positive and significant association with GI, ABN, DS and SDW.

Germination percentage was also well associated with seedling vigour index. This implies that increase in any of these attributes will lead to increase in other (Adebisi, 2008). Positive and significant association has been reported among seed quality characters in different crop species (Adebisi *et al.*, 2013; Idoko and Sabo 2014). From this study the negative and significant association between 100SWT and germination percentage, RLT; SHLT and SVI implies an increase in 100SWT results in a decrease in seed

quality attributes. The principal component analysis in this study revealed that the seed quality attributes such as germination percentage, germination index, seedling vigour index, shoot length, root length and seedling dry weight contribute significantly to the variation within the 22 genotypes of cowpea evaluated. According to Adebisi (2005), such attributes should be given due consideration in seed quality improvement programme for such crop species. Kehinde *et al.* (2005) and Adebisi (2008) reported that the first three principal components were the most important in reflecting the variation patterns among genotypes and the characters highly associated with these should be used in differentiating genotypes. The cluster analysis for seed quality attributes included in this study placed cowpea genotypes into four clusters with sub clusters for each, except cluster four with only three genotypes of one accession, one local and one improved, respectively. Clusters were grouped according to their different seed quality attributes among them. This result is in agreement with the findings of Sadia *et al.* (2012) who reported grouping of cultivars based on morphological differences rather than origin (source). All the clusters together with their sub clusters grouping included mixed genotypes of 22 cowpea with 11 accessions, 5 local and 6 improved. This indicates that they consisted of the heterogeneous group of genotypes with different source.

Conclusion

Significant differences occurred among the twenty-two cowpea genotypes for all the laboratory seed quality attributes evaluated. The mean performance of laboratory seed quality attributes revealed that NGB07614, Abewere, 150-Ex and Modupe were outstanding in term of seed quality attributes. All these outstanding genotypes fall within cluster three of the dendogram. This shows that selection for superior seed quality traits is possible among these cowpea accessions.

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Competing interests

The authors declare that they have no competing interests

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