

Research Article

Evaluation of 93 Accessions of African Yam Bean (*Sphenostylis stenocarpa*) Grown in Ethiopia for Physical, Nutritional, Antinutritional, and Cooking Properties

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African yam bean has immense food and nutrition potential and is resilient to adverse environmental conditions. Despite its potential, the crop is underutilized, which could be attributed to seed hardness (requiring about 6–24 hours of cooking time); and the abundance of antinutrient factors (tannin, phytate, and oxalate). This study evaluated the physical (seed hardness, cooking time) and chemical compositions (crude protein, tannin, phytate, and oxalate) of 93 AYB accessions grown in Ethiopia. The seed hardness of each accession was determined by the compression force and compression time using Texture Analyzer, whereas cooking time was ascertained using Mattson Bean Cooker. The accession's crude protein level, tannin, oxalate, and phytate were investigated from flour samples using standard laboratory procedures. Highly significant ($P < 0.01$) differences were observed for cluster means of compression force, cooking time, and oxalate. The accessions were grouped into three clusters: cluster-II was prominent with 42 accessions, while cluster-I had the least (25). The mean values for compression force ranged from $50.05 N \pm 10.25$ (TSs-423) to $278.05 N \pm 13.42$ (TSs-378) whereas compression time varied from $0.35 \text{ secs} \pm 0.02$ (TSs-334) to $5.57 \text{ secs} \pm 6.12$ (TSs-62B). Cooking time ranged from $127.50 \text{ mins} \pm 2.12$ (TSs-82A) to $199.50 \text{ mins} \pm 10.61$ (TSs-138B); crude protein ranged from $15.41\% \pm 0.11$ (TSs-269) to $24.51\% \pm 0.22$ (TSs-446). Tannin ranged from $0.61 \text{ mg/g} \pm 0.02$ (TSs-47) to $9.62 \text{ mg/g} \pm 0.03$ (TSs-334) likewise, phytate ranged from 0.28 ± 0.01 (TSs-137) to 7.01 ± 0.10 (TSs-3). Accessions TSs-55; TSs-82 showed the lowest oxalate content of $0.21\% \pm 0.01$; $0.21\% \pm 0.00$, respectively. Similarly, TSs-352; TSs-47 revealed the most abundant tannin content of 0.70 ± 0.00 and 0.70 ± 0.07 . The correlation analysis revealed a low positive and significant ($P < 0.05$) association ($r = 0.24$) between protein and phytate content.

1. Introduction

African yam bean (AYB) (*Sphenostylis stenocarpa*) is an underutilized legume crop of African origin. Several hypotheses proposed the crop's centre of domestication to include Ethiopia [1]; more so, the crop is mainly grown

across West Africa [2, 3], East, and Central Africa [3]. The crop produces seeds in pods and tuberous roots in some germplasm, of which its cultivation is associated with small-scale farmers [4–6].

AYB, which is considerably rich in protein, well-adapted, and locally available, can provide an additional protein

source and contribute to diet diversification in households in Sub-Saharan Africa. The protein content in AYB seeds is about 19 to 37% [6, 7]; in tubers, it is about 11–19% [8]. In addition, the seeds are rich in carbohydrates [9, 10], dietary fibre [8, 11], calcium, magnesium, and potassium [3, 10, 12]. The seeds are likewise used to prepare exceptional cuisine in some African communities. It could be boiled, roasted, fried, or steamed in combination with other ingredients. In addition, AYB seeds are used as a seasoning in soups [13, 14]. Also, AYB seeds were reportedly used to fortify food products, including breakfast meals, biscuits, and cereal flour [15–17]. AYB seeds are likewise crucial in the nutrient enrichment of animal feeds when used solely or combined with nutrient supplements [6, 18]. However, despite its immense potential for food, nutrition, and resilience to adverse environmental conditions, AYB is neglected, which could be attributed to constraints, including seed hardness [19–21], prolonged cooking time [6, 22, 23], and abundance of antinutrient factors (tannin, phytate, and oxalate) [5, 7, 10].

Although protein is essential for a healthy balanced diet, its consumption in several households in Africa is below the recommended level [24]. On the other hand, antinutritional factors affect nutrient uptake or availability [3, 25, 26]; however, some are beneficial when taken optimally [27]. Furthermore, seed hardness and cooking time are essential quality parameters; seed hardness significantly contributes to prolonged cooking time. The trait is also associated with delayed germination and provides good protection to seed pathogens [28]. In the same manner, long cooking time affects energy consumption, cost, and consumer preferences [29].

Characterizing germplasm for physical (seed hardness, cooking time) and chemical composition (protein, tannin, oxalate, and phytate) is necessary for the crop's utilization in diets and food fortification. Previously, the cooking time of AYB germplasm grown in Nigeria was evaluated using the boiling method, a nonrecommended approach [30–32]. Similarly, the protein, tannin, phytate, and oxalate content of about 50 AYB accessions cultivated in Nigeria were reported [5, 7, 8].

To the best of our knowledge, no published document is available on seed hardness and cooking time of AYB germplasm using recommended equipment such as Texture Analyzer and Mattson Bean Cooker. Additionally, even though the nutrient and antinutrient content of crops are affected by the environment in which they are grown and the target germplasm [33, 34], the nutrient (protein) and antinutrient (tannin, phytate, and oxalate) content of AYB germplasm cultivated under Ethiopia's environmental condition has not been reported.

The present research characterized and profiled the seed hardness and cooking time of 93 AYB germplasm cultivated in Ethiopia using Texture Analyzer and Mattson Bean Cooker, respectively. In addition, the accession's protein, tannin, phytate, and oxalate level were likewise investigated using standard laboratory procedures. Findings from the present study could provide crucial information on genotypes that can be selected for direct production or as parental

lines for use by breeding programs to improve the crop, enhancing vast production, and utilization of the crop for food and nutrition.

2. Materials and Methods

2.1. Experimental Site. The 93 AYB accessions used in the present study were sourced from the International Institute of Tropical Agriculture (IITA), Ibadan. The accessions were cultivated at Jimma Agricultural Research Center, Ethiopia, during the 2019/2020 cropping season between March and January. The location of the experimental site (1739 masl, N7°39.962', and E36°46.749') falls within the mid-altitude subhumid zone of Southwestern Ethiopia, in Oromia regional state. The maximum and minimum air temperatures of Jimma are 26.3 and 11.6°C, respectively. Jimma's annual precipitation falls within 1561 mm. The soil type of the experimental site is nitisol, reddish-brown with a loamy clay texture, and is slightly acidic [35].

2.2. Genetic Materials. The 93 accessions were morphologically characterized [36]. The physical properties of the accessions used in the present study are shown in Supplementary Table 1. The characterized accessions were packaged into three sets using standard procedures; on average, each accession weighed 19.44 grams. The first set of seeds (93 accessions) was submitted to the Postharvest Management Laboratory of Jimma University, Ethiopia, for the seed hardness test. The second set of 93 accessions was shipped to Melkassa Agriculture Research Center, Ethiopia, for cooking time profiling. Finally, the third set was sent to the Nutrition Sciences Laboratory, IITA, Nigeria, for protein and antinutrient analysis. The seed hardness, cooking test, protein, and antinutrient analysis were conducted in duplicate.

2.3. Seed Hardness Evaluation. The seed samples were dried in an oven at 37°C for 24 hours to maintain the air humidity across all seeds. Then, a texture analyzer (Stable Micro Systems TA.TX.Plus) was used to test the seed hardness of six randomly selected seeds from each accession using a Φ 5 mm probe. The instrument was calibrated with a load of 2 kg. The maximum compression force in Newton (Fmax/N) required to compress seeds was determined as seed hardness Song et al. [37]. The compression force and the compression time needed to break the seeds in seconds were obtained using the force-time curve.

2.4. Cooking Time Evaluation. Thirty-seed sampled from each accession were soaked for 24 hours in distilled water at room temperature. The automated Mattson Bean Cooker apparatus (Figure 1) was used to determine each sample's cooking time using the method described by Wang and Daun [38]. The Mattson Bean Cooker comprises of a cooking rack with 25 hollow saddles [38]. Out of the soaked seeds, 25 were selected at random and placed into each of the 25 saddles of the rack so that each plunger's tip rests on the seed's surface. The cooking is preceded by positioning the

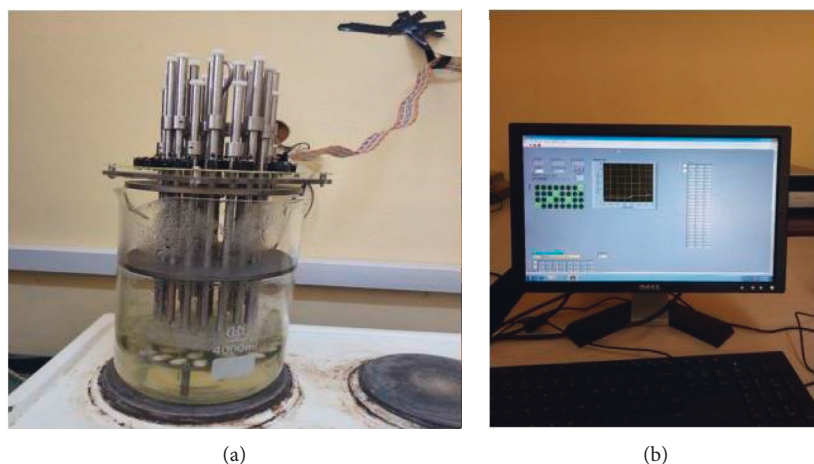


FIGURE 1: Schematic representation of automated Mattson bean cooker used in the present study: (a) shows the plungers, beaker, and the cooker and (b) shows the computer set up.

rack into a 2-L metal beaker containing 1.2 L of boiling water. When the seed becomes tender, the plunger penetrates the seeds and drops a short distance through the saddle hole. The time taken for each plunger to drop is automatically recorded, and the cooking time is defined as the penetration of 80% of the samples by the plunger [38, 39].

2.5. Nutrient and Antinutrient Analysis. Seeds devoid of impurities were crushed into a fine powder and packed in properly labelled airtight containers. Before analysis, the prepared samples were kept at 4–6°C. The investigation was carried out using standard laboratory procedures reported by Alamu et al. [40].

2.5.1. Determination of Crude Protein. The crude protein content was determined according to the Association of Official Analytical Chemists (AOAC) [41] procedure. About 10 ml of concentrated H_2SO_4 and selenium catalyst was added to 0.2 g of AYB fine powder. The mixture was heated continuously for 60–90 minutes on a digestion block operating at 420°C. The ammonium sulfate was distilled into a boric acid receiver solution and titrated with standard hydrochloric acid to obtain the total nitrogen. The amount of crude protein in the sample was calculated by multiplying the % nitrogen with the conversion factor of 6.25.

2.5.2. Determination of Tannin. Tannin was extracted using Folin-Dennis colorimetric method as described by Adegunwa et al. [42]. About 75 ml of water was added to 0.5 g fine AYB flour. The mixture was allowed to boil for 30 minutes, after which 1 ml of the solution was transferred into a volumetric flask, and 2 ml of a standard solution of tannic acid was added. Also, 0.5 ml of Folin Denis reagent and 1 ml of saturated Na_2CO_3 solution were added to the mixture. The total mix was made up to 10 ml with distilled water and incubated at room temperature for 30 minutes.

The absorbance was determined at 760 nm using a spectrophotometer. The total tannic acid content was expressed as mg of tannic acid equivalent per Gram of the extract.

2.5.3. Determination of Phytic Acid. Phytic acid was determined using the method described by Wheeler and Ferrel [43] with slight modifications. About 5 g of ground AYB sample was weighed into a conical flask, and 50 ml trichloroacetic acid (TCA) (3 g/100 g) was added. The solution was appropriately mixed using a mechanical shaker. An aliquot (10 ml) of the supernatant was transferred into a conical flask, and 4 ml $FeCl_3$ solution was added. The mixture was heated in a boiling water bath for 45 min, after which it was centrifuged at 3500 rpm for 15 min, and the supernatant carefully decanted. The precipitate was washed twice by dispersing in 25 ml of TCA and heating in a boiling water bath for 5. The cleaned precipitate was dispersed in 3 ml of water, and 3 ml of 1.5 N NaOH was added and adequately mixed. The mixture was topped up with distilled water to about 30 ml and was allowed to boil in a water bath for 30 minutes. The suspension (hot) was filtered using Whatman No. 1 filter paper, and the precipitate was washed with 60 ml of hot water. The cleaned precipitate was dissolved with 40 ml hot 3.2 N HNO_3 ; 5 ml of the suspension was transferred into a 100 ml volumetric flask, and 20 ml of 1.5 M KSCN was added. The total volume was made up to 100 ml with distilled water, and the colour absorbance was immediately read at 480 nm. The iron content was calculated from a $Fe(NO_3)_3$ standard curve, and the phytate phosphorus was calculated from the iron results assuming a 4 : 6 iron : phosphorus molecular ratio.

2.5.4. Determination of Oxalate. Oxalic acid was determined using a colorimetric method described by Bergerman and Elliot [44]. About 0.5 g of powdered AYB sample was weighed into a 50-ml volumetric flask, and 30 ml of 0.25 N HCl was added. After boiling for 15 min, the solution was allowed to cool at room temperature, and the volume was

made up with 0.25 N HCl. The above solution was used as the extract oxalic acid determination. Indole reagent was freshly prepared by dissolving 100 mg of indole in 100 ml of concentrated sulfuric acid. The assay mixture contained 2 ml of standard oxalic acid solution with concentrations ranging from 0.100 to 1.00 mg per ml prepared in 1 N H₂SO₄. About 2 ml of 1 N sulfuric acid was used as the blank solution. In each test tube, 2 ml of indole reagent was added, and the test tubes were placed in a water bath at 80–90°C for 45 minutes. The tubes were kept at room temperature, and absorbance was measured at 525 nm on a spectrophotometer.

2.6. Statistical Analysis. All data analyses were carried out using the *R* statistical package version 4.1.1 [45]. Descriptive statistics for all the parameters were generated, and the analysis of variance (ANOVA) was performed using the *Agricola R* package. The *hclust* function was used to construct the dendrogram. The *Chart.correlation* function from the *PerformAnalytics* package generated the correlation plots. The principal component analysis (PCA) and PCA biplot were generated using the *FactorMineR* package.

3. Results

3.1. Descriptive Statistics of Evaluated Parameters. Table 1 shows the descriptive statistics for compression force, compression time, cooking time, crude protein, tannin, phytate, and oxalate across the studied 93 AYB accessions. The maximum compression force required to break a seed was 278.05 N ± 13.42, while the least force needed was 50.05 N ± 10.25. Furthermore, the highest time spent compressing a seed was 5.57 secs ± 6.12, whereas the least time taken was 0.35 secs ± 0.02. Also, the maximum time spent to cook an accession was 199.50 mins ± 10.61, while the least time required was 127.50 mins ± 2.12. The accessions presented the mean values of 177.59 N, 1.18 secs, and 159.95 mins for compression force, compression time, and cooking time, respectively. The accession's crude protein content varied from 15.41% ± 0.11 to 24.51% ± 0.22 with a mean value of 19.95%; while tannin content ranged from 0.61 mg/g ± 0.02 to 9.62 mg/g ± 0.03 with a mean of 4.71 mg/g. Phytate content ranged from 0.28% ± 0.01 to 7.01% ± 0.10 whereas oxalate content was between 0.21% ± 0.01 to 0.70% ± 0.07.

3.2. Hierarchical Cluster Analysis of the AYB Accessions. The hierarchical cluster analysis performed on 93 accessions considering the following variables: compression force, compression time, cooking time, crude protein, tannin, phytate, and oxalate, revealed three clusters (Figure 2). Cluster-II was the most prominent consisting of 42 accessions that accounted for 45.2% of the total germplasm; similarly, cluster-III followed with 26 accessions accounting for 28.0% of the studied accessions. The total number of accessions in cluster-I was 25. Table 2 shows the hierarchical cluster details.

3.3. Analysis of Variance for the Clusters. Table 3 presents the analysis of variance of the physical and chemical composition of 93 AYB accessions by hierarchical clusters. Highly significant differences ($P < 0.01$) were found among the clusters for compression force, cooking time, and oxalate while significant difference was found for tannin ($P < 0.05$). Accessions grouped in clusters-I (157.00 N), required the least compression force, while members of cluster-II (164.00 N) and III (218.00 N), were characterized by a much higher compression force. Cluster-III was significantly different from the clusters I and II. Cluster-III was attributed with the highest compression time (1.32 secs); the cluster was not significantly different from clusters-I (1.04 secs) and II (1.18 secs). The shortest cooking time (142.00 mins) across cluster was associated with accessions grouped in cluster-I; the cluster was significantly different from clusters-II (162.00 mins) and III (174.00 mins).

Considering crude protein, accessions in cluster-II had the highest protein content of 20.20%. The protein content of accessions in the cluster (III) was not significantly different from the amount in Clusters-I (19.90%) and III (19.70%). The tannin content (5.29 mg/g) obtained from accessions in cluster-III was higher than the values in the other clusters. The content was significantly different ($P < 0.05$) from the values in cluster-I (3.96 mg/g) but showed no difference with content in cluster-II (4.80 mg/g). Furthermore, no significant differences were observed among the clusters for phytate; however, for oxalate, highly significant difference ($P < 0.01$) was observed among the cluster. The least oxalate content was found in cluster-I (0.33%); the cluster showed significant difference with oxalate content of accessions in cluster-II.

3.4. Crude Protein Composition of AYB Accessions. The list of AYB accessions with the top 20 protein contents is presented in Table 4. The hierarchical cluster representation for the top 20 high protein accessions revealed that cluster-I had 4 accessions, whereas cluster-II and cluster-III had 11 and 5 accessions. Out of the 20 accessions with high protein content, 13 had brown seed colour, and the majority were grouped in cluster-II. A brown-black seeded accession (TSs-446) grouped in cluster-I presented the highest protein content of 24.51%. The protein content of the accession was 19.50% higher than the average mean (19.73%) reported for the studied materials. TSs-446 was closely followed by a black seeded accession (TSs-13) with a protein value of 24.49%, the accession grouped into cluster-II. The protein content (23.95%) obtained in TSs-448 ranked as the third top; the value was higher than the average mean of all the accessions by 17.62%. The accession (TSs-448) had brown seed colour and was grouped with 5 other accessions in cluster-II. The protein content in TSs-423 was 23.89% and the accession was found in cluster-II. TSs-443 identified as the accession with the fifth protein abundance with a mean value of 23.77%, grouped in cluster-II had a protein value of 17.05% higher than the grand mean. Additional accessions that showed high protein content include grey seeded TSs-119, brown seeded TSs-197, brown seeded TSs-58, brown seeded TSs-155, and brown seeded TSs-7 with

TABLE 1: Descriptive statistics of 93 AYB accessions for compression force, compression time, cooking time, crude protein, tannin, phytate, and oxalate.

Descriptive statistics	Compression force (N)	Compression time (secs)	Cooking time (mins)	Crude protein (%)	Tannin (mg/g)	Phytate (%)	Oxalate (%)
NOB	93	93	93	93	93	93	93
Minimum value	50.05 ± 50.66	0.35 ± 0.02	127.50 ± 2.12	15.41 ± 0.11	0.61 ± 0.02	0.28 ± 0.01	0.21 ± 0.01
Maximum value	278.05 ± 13.42	5.57 ± 6.12	199.50 ± 10.61	24.51 ± 0.22	9.62 ± 0.03	7.01 ± 0.10	0.70 ± 0.07
Range	228.01	5.23	72.00	9.10	9.02	6.73	0.50
Median value	176.66	1.03	150.00	19.87	4.40	5.34	0.41
Mean value	177.59	1.18	159.95	19.95	4.71	5.16	0.45
SE	5.25	0.07	2.11	0.21	0.19	0.01	0.01
SD	50.66	0.72	20.31	1.99	1.80	0.93	0.14
CV	0.29	0.61	0.13	0.10	0.38	0.18	0.30

AYB, African yam bean; NOB, number of observations; SE, standard error of the mean; SD, standard deviation; CV, coefficient of variation. All values are means ± SD of duplicate determination.

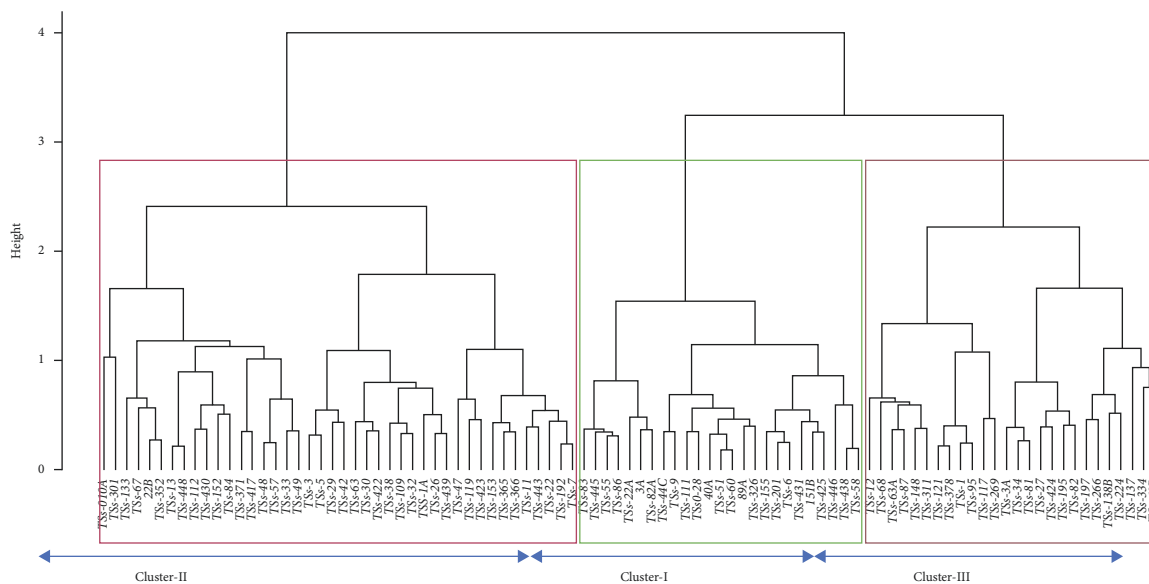


FIGURE 2: Hierarchical cluster dendrogram based on Gower distance matrix of physical and chemical traits. The dendrogram was created in R software version 4.1.1.

corresponding protein values of 23.48%, 23.43%, 22.74%, 22.49%, and 22.44%. The mean value of TSs-119 deviated from the average mean value of all the accessions by 12.09%. Likewise, the mean value obtained for TSs-7 was higher than the average mean of the total accessions by 11.10%. Other accessions that presented high protein content were TSs-371 (22.43%), TSs-334 (22.08%), TSs-365 (22.06%), TSs-11 (22.04%), TSs-82 (21.92%), TSs-57 (21.82%), TSs-201 (21.74%), TSs-424 (21.73%), TSs-48 (21.68%), and TSs-62B (21.52%).

3.5. Textural and Chemical Profiling of 93 AYB Accessions.

The textural and chemical profiling of 50 accessions are presented in Table 5. Supplementary Table S2 shows the textural and chemical profiling of 93 accessions. A sufficient level of phenotypic variability was observed for the investigated traits. TSs-334 presented a compression force (CF) of 105.53 N, compression time (CT) of 0.35 secs, and cooking time (CKT) of 168.00 mins. The accession (TSs-334) revealed

the most tannin (TA) content (9.62 mg/g); its corresponding value for phytate (PH) and oxalate (OX) 5.84% was 0.29%. Also, TSs-334 revealed the least compression time (0.35) across all the accessions. Furthermore, TSs-47 showed the following seed quality profile: compression force (177.95 N), compression time (0.81 secs), cooking time (142.50 mins), tannin (0.61 mg/g), and phytate (5.39 mg/g). The highest oxalate content (0.70%) was likewise reported in TSs-47. In addition, accession TSs-137 had the corresponding values: 211.69 N, 0.71 secs, 184.50 mins, 8.04 mg/g, and 0.41% for compression force, compression time, cooking time, and oxalate, respectively. The accession (TSs-137) reported negligible phytate content (0.28%) compared with values reported for other accessions. On the contrary, the highest phytate level (7.01%) was obtained in TSs-3; the following profile was likewise documented in accession TSs-3; compression force, 145.67 N; compression time, 1.53 secs; cooking time, 145.50; tannin, 3.64 mg/g, and oxalate 0.58%. In oxalate analysis, the oxalate presentation was lowest (0.21%) in TSs-55; the values 183.64 N, 0.59 secs, 145.50 mins, 3.05 mg/g, and

TABLE 2: Cluster analysis of 93 AYB accessions.

Cluster Object	I 25	II 42	III 26
	151B	22B	TSs-1
	3A	TSs-109	TSs-117
	40A	TSs-10A	TSs-12
	89A	TSs-11	TSs-121
	TSs-111	TSs-112	TSs-137
	TSs-155	TSs-119	TSs-138B
	TSs-201	TSs-13	TSs-148
	TSs-22A	TSs-133	TSs-195
	TSs-28	TSs-152	TSs-197
	TSs-326	TSs-153	TSs-224
	TSs-425	TSs-192	TSs-266
	TSs-431	TSS-1A	TSs-269
	TSs-438	TSs-22	TSs-27
	TSs-445	TSs-26	TSs-311
	TSs-446	TSs-29	TSs-334
	TSs-44C	TSs-3	TSs-34
	TSs-51	TSs-30	TSs-378
	TSs-55	TSs-301	TSs-3A
	TSs-58	TSs-32	TSs-424
	TSs-6	TSs-33	TSs-62B
	TSs-60	TSs-352	TSs-63A
	TSs-82A	TSs-365	TSs-68
	TSs-83	TSs-366	TSs-81
	TSs-86	TSs-371	TSs-82
	TSs-9	TSs-38	TSs-87
		TSs-417	TSs-95
		TSs-42	
		TSs-422	
		TSs-423	
		TSs-430	
		TSs-439	
		TSs-443	
		TSs-448	
		TSs-47	
		TSs-48	
		TSs-49	
		TSs-5	
		TSs-57	
		TSs-63	
		TSs-67	
		TSs-7	
		TSs-84	

AYB, African yam bean.

5.36% were, respectively, detected for compression force, compression time, cooking time, tannin, and phytate in TSs-55. TSs-352 was another accession with a high oxalate value (0.70%). The compression force of the accession was 170.91 N; compression time was 1.61 secs, cooking time was 166.50 mins, its tannin level was 7.63 mg/g, and phytate 4.97%. The analysis of the cooking test showed TSs-138B as the longest cooking accession; the accession required an average cooking time of 199.50 mins which is contrary to the average time (127.50 mins) needed to achieve tenderness in TSs-82A. TSs-138B had corresponding values of 239.41 N, 2.44 secs, 6.78 mg/g, 5.88%, and 0.45% for compression force, compression time, tannin, and oxalate, respectively. In the texture analysis, TSs-378 was revealed as the accession with the toughest seed texture; the accession presented the highest

compression force (278.05 N) and the following values 0.75 secs, 147.00 mins, 6.16 mg/g, 6.05%, 0.41%, respectively, for compression time, cooking time, tannin, phytate, and oxalate. Contrarily, the compression force of 50.05 N was needed to crush seeds obtained from accession TSs-423; the compression force (50.05 N) was the minimum value observed across the 96 accessions. The seed quality profile of accession TSs-62B is as follows; compression force (188.90 N), compression time (5.57 secs), cooking time (168.00 mins), tannin (4.52 mg/g), phytate (4.71%), and oxalate (0.30%). The compression time spent when crushing TSs-62B was the maximum (5.57 secs) reported for all the accessions. The mean values of 216.34 N, 0.71 secs, 184.50 mins, 4.26 mg/g, 5.27%, 0.49% were reported in compression force, compression time, cooking time, tannin, phytate, and oxalate, respectively, for accession TSs-430.

3.6. Correlation Analysis of Protein, Antinutrients, Cooking Time, Compression Force, And Compression Time. The correlation analysis between protein, antinutrients, cooking time, compression force, and compression time investigated across 93 accessions (Figure 3). A positive and significant ($P < 0.05$) correlation (0.24*) was found between protein and phytate; however, a nonsignificant relationship was found among the other parameters. Protein and tannin showed a weak and positive (0.04) relationship, whereas the association between protein and oxalate was 0.01.

3.7. Principal Component Analysis (PCA). Principal component (PC) analysis was computed to determine the most discriminative variables on the 93 AYB accessions. The principal components values ≥ 0.40 are presented in Table 6. The first five principal components (PC) axis cumulatively contributed 79.2% of the observed variations with an eigenvalue of 5.53. PC 1 to PC 3 showed eigenvalues higher than one and contributed 52.5% of the total variations in the studied germplasm. Figure 4 presents the first two PC axis and the percentage contribution of each trait to the axis. PC axis 1 contributed 19.9% of the total variation, whereas PC axis 2 contributed 17.3%. The traits that contributed to PC1 were crude protein, phytate, and compression force; however crude protein (-0.55) and phytate had a negative loading (-0.55). PC axis 2 contributed 17.3% of the total variations across the traits. The accessions that contributed much of the observed variation in PC2 were tannin (0.56), cooking time (0.59), and compression time (0.40) (Table 6). Oxalate (0.73) and compression time (-0.46) were the traits with high loadings in PC3. Furthermore, tannin (0.60), cooking time (-0.40) and compression time (-0.57) had high loadings in PC 4. Crude protein (0.42), cooking time (0.58), and compression time (-0.52) were the traits that contributed most of the variation observed in PC 5 (Table 6).

3.8. PCA Biplot. The PCA biplot grouped the studied accessions (Figure 4) into three clusters. Cluster-I had 25 accessions, out of which 11 were of Nigerian origin, and the other 14 were accessions whose origins were not available.

TABLE 3: Analysis of variance of the physical and chemical composition of 93 AYB accessions by hierarchical cluster.

Cluster	CF (N)	CT (secs)	Cooking time (mins)	Crude protein (%)	Tannin (mg/g)	Phytate (%)	Oxalate (%)
I	157.00b	1.04a	142.00c	19.90a	3.96b	5.16ab	0.33b
II	164.00b	1.18a	162.00b	20.20a	4.80ab	5.37a	0.59a
III	218.00a	1.32a	174.00a	19.70a	5.29a	4.83b	0.35b
Mean	177.59 ± 50.66	1.18 ± 0.72	159.95 ± 20.31	19.95 ± 1.99	4.71 ± 1.80	5.16 ± 0.93	0.45 ± 0.14
<i>P</i> value	<0.01	0.40	<0.01	0.60	<0.05	0.07	< 0.01
CV	24.90	61.20	10.40	10.10	37.00	17.70	12.10

AYB, African yam bean; CF, compression force; CT, compression time; CV, coefficient of variation. Mean values with different letters along the same column are significantly different ($P < 0.05$).

TABLE 4: Crude protein content, seed color of 20 AYB accessions by hierarchical clusters.

Accessions	Crude protein (%)	Cluster	Seed color
TSs-446	24.51 ± 0.22	I	Brown-black
TSs-13	24.49 ± 0.21	II	Black
TSs-448	23.95 ± 0.02	II	Brown
TSs-423	23.89 ± 0.20	II	Brown
TSs-443	23.77 ± 0.01	II	Brown
TSs-119	23.48 ± 0.34	II	Grey
TSs-197	23.43 ± 0.11	III	Brown
TSs-58	22.74 ± 0.24	I	Brown
TSs-155	22.49 ± 0.21	I	Brown
TSs-7	22.44 ± 0.40	II	Brown
TSs-371	22.43 ± 0.05	II	Brown
TSs-334	22.08 ± 0.16	III	Brown
TSs-365	22.06 ± 2.71	II	Brown
TSs-11	22.04 ± 0.06	II	Grey
TSs-82	21.92 ± 0.11	III	Grey
TSs-57	21.82 ± 0.59	II	Grey-black
TSs-201	21.74 ± 0.06	I	Brown
TSs-424	21.73 ± 0.17	III	Brown-black
TSs-48	21.68 ± 0.01	II	Brown
TSs-62B	21.52 ± 0.14	III	Brown

All values are means ± standard deviation of duplicate determination.

Cluster-II was associated with 42 accessions, of which more than half of the accessions were of Nigerian origin and 1 accession of Bangladeshi origin. A total of 26 accessions were grouped in cluster-III; the cluster had 1 accession of Ghana origin, 10 of Nigerian origin, and other accessions whose origins were not reported. The accessions in cluster-I were uniquely characterized by low protein, oxalate, phytate, reduced cooking time, compression force, and compression time whereas accessions in cluster-II were characterized by high protein, phytate, oxalate, compression time, and tannin content. However, accessions in cluster-III were characterized by high cooking time and compression force.

4. Discussion

Numerous attributes, including nutrient content, cooking time, and seed hardness, influence consumers' and processors' food choices [46, 47]. The observed variations revealed in crude protein composition, antinutrient factors, cooking time, compression force, and compression time could result from existing variations in the studied accessions. The variations were further justified by the significant differences ($P < 0.01$)

shown in cluster means of some of the accessions. The present findings agree with previous reports [48]. The authors reported highly significant differences in cluster means of nutritional and antinutrition factors of 20 AYB accessions. Likewise, Ndidi et al. [10] reported significant differences across proximate components evaluated for raw and processed AYB seeds.

Protein is an essential nutrient required for a healthy diet; however, several households across sub-Saharan Africa cannot meet the daily protein recommendations, resulting in health-related issues, including stunted growth [24]. Interestingly, AYB, a crop of African origin that is locally available and considerably rich in protein, could reduce protein deficiencies and promote dietary diversification in sub-Saharan Africa. The protein content (24.51%) obtained in this study is sufficiently high and is similar to the value (25.08%) reported for 25 accessions evaluated in Nigeria [7]. Similarly, the result corresponds with the value (25.60%) obtained across 40 accessions (27 from IITA and 13 from the Institute of Agricultural Research and Training (IAR&T) Nigeria) morphologically characterized in Nigeria for two seasons [49]. In this study, TSs-446 recorded the highest percentage of protein content and could be utilized by

TABLE 5: Textural and chemical profiling of 50 AYB accessions.

Accession	CF (N)	CT (sec)	CKT (min)	TA (mg/g)	PH (%)	OX (%)	Accession	CF (N)	CT (Sec)	CKT (Min)	TA (Mg/g)	PH (%)	OX (%)
TSs-334	105.53	0.35	168.00	9.62	5.84	0.29	TSs-26	222.12	0.77	139.50	2.61	5.34	0.58
TSs-47	177.95	0.81	142.50	0.61	5.39	0.70	TSs-1A	199.34	1.82	142.50	5.44	6.20	0.51
TSs-137	211.69	0.71	184.50	8.04	0.28	0.41	TSs-33	164.49	0.53	186.00	2.77	5.73	0.65
TSs-3	145.67	1.53	145.50	3.64	7.01	0.58	TSs-38	267.52	0.74	168.00	3.42	5.48	0.58
TSs-55	183.64	0.59	145.50	3.05	5.36	0.21	TSs-82	179.77	1.42	187.50	3.17	6.17	0.21
TSs-352	170.91	1.61	166.50	7.63	4.97	0.70	TSs-417	167.98	0.58	187.50	9.12	5.94	0.61
TSs-138B	239.41	2.44	199.50	6.78	5.88	0.45	TSs-371	216.05	0.61	189.00	8.12	5.26	0.64
TSs-82A	91.57	0.40	127.50	3.93	4.95	0.26	40A	180.63	0.98	139.50	2.13	5.77	0.40
TSs-378	278.05	0.75	147.00	6.16	6.05	0.41	TSs-84	114.08	0.92	189.00	3.49	4.99	0.53
TSs-423	50.05	1.03	139.50	4.96	5.25	0.60	TSs-83	160.84	0.58	139.50	4.16	3.30	0.31
TSs-62B	188.90	5.57	168.00	4.52	4.71	0.30	TSs-68	241.63	1.80	166.50	4.50	5.93	0.30
TSs-430	216.34	0.71	184.50	4.26	5.27	0.49	TSs-30	215.99	2.00	150.00	5.65	3.87	0.64
TSs-224	257.21	1.11	185.10	7.97	5.93	0.37	151B	134.11	1.35	151.50	7.19	5.86	0.36
TSs-311	221.58	0.60	184.50	5.20	5.55	0.35	TSs-148	242.59	0.68	190.50	4.11	4.43	0.38
TSs-109	220.79	1.16	166.50	3.71	5.11	0.58	TSs-63A	208.61	1.06	169.50	4.18	4.10	0.46
TSs-378	278.05	0.75	147.00	6.16	6.05	0.41	TSs-48	151.23	0.87	196.50	2.66	6.04	0.57
TSs-1	259.46	0.72	139.50	4.73	4.96	0.37	TSs-51	188.68	0.72	142.50	2.89	4.82	0.33
TSs-67	101.69	0.49	165.30	4.44	6.45	0.62	TSs-60	193.66	1.15	139.50	3.26	5.53	0.33
TSs-112	156.02	1.39	190.50	5.03	5.45	0.47	TSs-22A	100.81	0.38	148.50	4.04	3.48	0.33
TSs-11	204.39	1.40	138.00	4.38	6.02	0.59	TSs-9	149.76	1.02	141.30	3.04	4.10	0.40
TSs-326	156.18	0.51	150.00	4.77	5.00	0.29	TSs-95	276.30	0.81	142.50	5.76	4.64	0.36
TSs-133	129.55	3.45	168.00	5.76	5.17	0.60	TSs-44C	95.83	0.75	141.00	2.65	5.12	0.38
TSs-152	151.66	0.93	184.50	7.23	4.76	0.58	TSs-81	155.28	1.08	186.00	3.45	4.14	0.34
TSs-153	132.24	1.99	139.50	6.08	6.06	0.52	TSs-121	275.19	0.81	151.50	6.10	5.15	0.41
3A	90.64	1.24	139.50	1.89	5.13	0.345	TSs-266	258.75	2.09	190.50	4.26	5.69	0.31

CF, compression force; CT, compression time; CKT, cooking time; TA, tannin; PH, phytate; OX, oxalate. All values are means of two determinations.

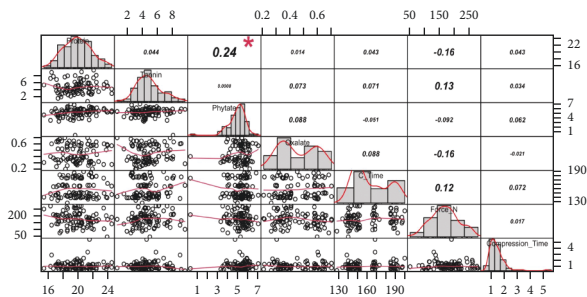


FIGURE 3: Correlation among physical and chemical traits evaluated in AYB accessions. The association of traits were studied using Pearson's correlation package in R software version 4.1.1. The x- and y-axis represent the respective scales of each variable under consideration. Below the diagram are histograms showing the distribution of the traits.

consumers and breeders for protein enrichment and further improvement, respectively.

Several researchers documented the presence of tannin, phytates, and oxalates in AYB accessions evaluated in Nigeria [7, 11, 21]. The tannin content (9.62 mg/g) revealed in the present research differs in quantity from the values of 0.12 mg/g and 3.34 mg/g previously reported [11, 50]. However, much higher tannin content (18.09 mg/g) and (38.80 mg/g) have been reported in AYB [7, 10]. Therefore, the accession (TSs-47) with the least tannin level could help develop AYB cultivars with reduced tannin content. More so, the phytate level (0.28%–7.01%) revealed in this study is higher than the content (0.002–0.72%) previously reported across AYB accessions [7, 10, 11]. The presence of high

tannin content in this research could result from genetic differences in the studied accessions and environmental conditions, which are factors known to influence nutrient quality in crops [34]. Nevertheless, phytate, which has been identified as a contributory factor to long cooking time in legumes [50], was minimal in TSs-137. Therefore, this accession (TSs-137) could be a choice material for developing cultivars with reduced cooking time.

The assessment of cooking time using Mattson Bean Cooker is one standard approach for the phenotypic evaluation of grain legumes [51]. In like manner, cooking time is linked to consumers' choice as consumers are more inclined to convenient foods than food with prolonged cooking time. Also, long cooking time limits the adequate acceptance of some legumes [47]. The long cooking time of 3–6 hours [52, 53] attributed to AYB limits the consumption of the crop. To our knowledge, this study is the first to use the Mattson Bean Cooker to assess the cooking time of AYB seeds. The range of cooking time (127.50–199.50 mins) recorded in this study further confirms the hard to cook attributes of AYB seeds. The cooking time obtained in this study is lower than the values (300 mins and above) previously reported using traditional cooking methods [53, 54].

Seed hardness is a complex quantitative trait influenced by the environment and genetic architecture of the material [55]. Although the texture and microstructure of AYB seeds were previously reported using a scanning electron microscope, the seed hardness analysis of AYB has not been reported. The present research is the first to use a texture analyzer for seed hardness tests in the crop. However, the

TABLE 6: Principal component (PC) analysis of textural and chemical properties of 93 AYB accessions.

Parameter	PC1	PC2	PC3	PC4	PC5
Crude protein	-0.55	—	—	—	0.42
Tannin	—	0.56	—	0.60	—
Phytate	-0.55	—	—	—	—
Oxalate	—	—	0.73	—	—
Cooking time	—	0.59	—	-0.40	0.58
Compression force	0.52	—	—	—	—
Compression time	—	0.40	-0.46	-0.57	-0.55
Eigenvalue	1.39	1.21	1.06	0.97	0.90
Percent variation	19.93	17.33	15.20	13.82	12.90
Cumulative variation	19.93	37.26	52.46	66.28	79.17

AYB, African yam bean. The PC represented value ≥ 0.40 .

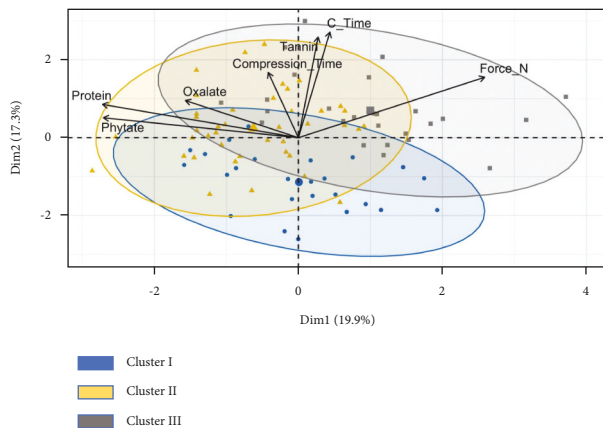


FIGURE 4: Principal component (PC) biplot showing the contribution of each trait to the variation observed in the studied African yam bean accessions. Force_N; compression force, CT_mins; cooking time, clus; cluster. The biplot was created using R software version 4.1.1.

texture analyzer approach has been reported in food legumes [28, 39, 55]. Therefore, out of the 93 accessions evaluated for seed hardness in this study, the extreme accessions (TSs-378 and TSs-423) identified could serve as parental materials to improve seed hardness in AYB.

The positive relationship between protein and phytate suggests that phytate can affect protein availability. Ndidi et al. [10] reported an influence of phytate on the protein content of processed AYB seeds. The high PC loading for crude protein, phytate, oxalate, compression time, and compression force content suggests the relevance of the traits in distinguishing the studied accessions. Ajibade et al. [48] also reported oxalate, tannin, and phytate as important discriminative traits in AYB evaluation. Finding from the PCA biplot shows that accessions in cluster-II are promising materials for improving protein, phytate, and oxalate content. Also, accessions in cluster-I could help develop cultivars with reduced cooking time and low antinutritional content.

5. Conclusion

The use of Mattson Bean Cooker and Texture Analyzer in AYB cooking test and seed hardness analysis effectively evaluated AYB seed's quality. The fast-cooking and soft

seeded accessions revealed in the present study could positively impact the utilization of the crop. The study also successfully identified germplasm with high protein levels and accessions with either high or low antinutrient content. The extent of antinutrients reported in this study should not be negatively perceived because antinutrient can also be exploited for possible health benefits. Nevertheless, most legumes are processed before they can be consumed, and several methods, including soaking, sprouting, and boiling, have shown effectiveness in reducing the amount of antinutrients in food. Above all else, the studied accessions have a high potential to address identified limitations in AYB. A molecular approach to identify the evaluated trait's genetic architecture should be considered for further study.

Data Availability

All data generated to support the findings of this study are included in the paper.

Conflicts of Interest

The authors declare no conflicts of interest.

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Supplementary Materials

Supplementary Table S1: physical properties of the 93 AYB accessions used in the present study. Supplementary Table S2: phenotypic variations of compression force, compression time, cooking time, protein, tannin, and phytate across 93 AYB accessions. (*Supplementary Materials*)

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