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Original Research Article

Genetic Potential and Phytochemical Diversity of African Yam Bean (*Sphenostylis stenocarpa*): A Gateway to Nutritional Security and Crop Improvement

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ABSRTACT

This study examines the genotypic and phytochemical diversity of African yam bean (*Sphenostylis stenocarpa*), an underutilised legume with potential applications in sustainable agriculture and nutrition. Phytochemical profiles of seeds from five distinct landraces, sourced from Cross River, Abia, Benue, Ogun, and Niger States in Nigeria, were analysed using qualitative and quantitative methods. The qualitative analysis revealed significant variability in phytochemical compounds, including alkaloids, flavonoids, saponins, tannins, and terpenoids, across the landraces. Alkaloids were consistently present in moderate concentrations across all accessions, suggesting genetic stability in their synthesis. Conversely, flavonoid levels were highest in the Benue landrace, indicating potential region-specific environmental or genetic factors influencing their production. Saponins were most abundant in the Cross River and Abia landraces, while tannins were uniformly high, with the Benue landrace showing the greatest concentration. Terpenoid levels were generally low but reached moderate peaks in the Niger landrace. Quantitative analysis confirmed these patterns but found no significant differences (P> 0.05) in phytochemical concentrations among the landraces, highlighting a general consistency in their distribution. The findings emphasise the nutritional and agronomic potential of African yam beans, particularly the Benue and Cross River landraces, which demonstrate promising phytochemical profiles. This research underscores the importance of conserving African yam bean genetic diversity and utilising its phytochemical profiles. This research underscores the importance of conserving African yam bean genetic diversity and utilising its phytochemical traits for breeding programmes aimed at enhancing food security, addressing malnutrition, and exploring industrial applications.

Keywords: African Yam Bean, Phytochemical Diversity, Crop Improvement, Nutritional Security, Landrace variability.

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Introduction

African yam bean (Sphenostylis stenocarpa), native to Abyssinia (modern-day Ethiopia), is predominantly found in the wild but is widely cultivated across West Africa, particularly in countries such as Nigeria, Côte d'Ivoire, Cameroon, Togo, and Ghana.^{1,2} Its cultivation is expanding into East, Central, and Southern Africa due to its highly protein-rich seeds and tubers, especially in subsistence farming communities where it serves as a reliable food source 34.5 Despite its nutritional value, the crop faces the risk of extinction due to a lack of economic support for its cultivation.⁶ Commonly referred to as "African yam bean" (AYB), the plant is valued for its edible tubers.⁷ This annual climbing legume is often intercropped with yam (Dioscorea spp.) in many regions, sharing the same stakes.8 Roots and tubers of AYB contribute greatly to the culinary traditions of several tropical and subtropical communities.^{1,9,10} Among the seven species within the Sphenostylis genus, AYB holds the greatest economic importance in Africa 11.

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It is particularly prized for its seeds in countries like Nigeria, Ghana, Côte d'Ivoire, Togo, and Cameroon, while in other regions such as Gabon, the Democratic Republic of Congo, Ethiopia, Malawi, and Zimbabwe, it is deliberately cultivated for its tubers.^{12,13}

The AYB is an exceptionally nutritious crop with numerous health benefits. Its seeds are an excellent protein source, containing 19-25% protein and essential amino acids such as lysine, making it particularly effective in addressing protein deficiencies, especially in areas with limited access to animal protein sources, 9,10,14,15,16,17 The tubers are rich in carbohydrates and provide a substantial energy source, making the crop dual-purpose by addressing both energy and protein requirements. This versatility highlights its potential as a sustainable food crop for meeting nutritional and energy needs.^{3,4,9,18,19,20} Furthermore, both the seeds and tubers are abundant in dietary fiber, which supports digestion, enhances gut health, and prevents constipation.^{4,9,10,21} This crop is also a rich source of essential micronutrients, including calcium, potassium, magnesium, and iron, which contribute to bone health, blood pressure regulation, and anemia prevention. It also contains B vitamins such as thiamine and riboflavin, which are vital for energy metabolism and overall physiological function.²² The seeds also have a relatively low fat content, making them suitable for low-fat diets while still providing essential fatty acids required for maintaining metabolic health.9

Beyond its impressive nutrient profile, the African yam bean (AYB) is recognised for its phytochemicals and bioactive compounds, which offer health benefits such as reducing the risk of lifestyle-related diseases.^{23,24,25,26,27} Phytochemicals are naturally occurring secondary metabolites found in plants, fruits, and seeds, often found in smaller quantities than primary metabolites.²⁸ These bioactive compounds have been linked to a lower likelihood of conditions like heart disease and degenerative conditions caused by oxidative stress. Oxidative stress arises from an imbalance in the body's antioxidant defenses, leading to cellular damage.^{24,29,30} Antioxidant-rich phytochemicals in AYB help

neutralize harmful free radicals, thereby mitigating oxidative damage and promoting overall health. 31

A high phytochemical content is indicative of significant antioxidant activities, including free radical scavenging potential,^{27,29,30,32} and metal-chelating effects.²⁷ These antioxidant properties are crucial in neutralising harmful free radicals, thereby reducing oxidative stress and its associated risks. Furthermore, Ibiyinka *et al.* (2014) ³³ reported that compounds such as carotenoids, vitamins, and volatile oils found in the African yam bean further enhance its antioxidant activity. This makes AYB a valuable dietary inclusion for promoting health and mitigating oxidative damage. Increasing its consumption is therefore recommended to maximise these health benefits.

Exploring the genetic basis of underutilised crops like the African vam bean (AYB) is essential for unlocking their potential to improve food security and tackle malnutrition. Comprehensive assessments of genetic diversity, encompassing cultivated varieties and landraces, are fundamental for understanding the genetic structure of AYB. These evaluations should utilise diverse methodologies, including morphological, biochemical, cytogenetic, molecular, and phytochemical analyses.³⁴ Understanding the genetic variation within AYB is vital for breeding programmes aimed at enhancing the crop's productivity and resilience. Effective breeding requires a thorough understanding of genetic diversity, which also serves as the basis for conserving and utilising plant genetic resources in sustainable agricultural practices.^{35,36} Traditional genetic assessment methods, such as morphological and biochemical markers have limitations, prompting the adoption of phytochemical-based markers. These compounds are increasingly used for plant identification and classification due to their species-specific nature, making them reliable tools for distinguishing between plant species. Moreover, phytochemical markers provide valuable insights into a plant's medicinal and economic potential. 37,38,39 By leveraging phytochemical markers, researchers can enhance the precision of genetic analyses, improve conservation strategies, and develop targeted breeding programmes for AYB. Such advancements have the potential to transform AYB into a more widely cultivated crop, addressing global challenges of food security and malnutrition. Progress in phytochemical research has been facilitated by the development of rapid and accurate techniques for screening plants for specific chemicals. Methods such as chromatography (paper and thin layer) and electrophoresis have greatly enhanced the ability to identify and quantify phytochemicals in plant species.^{1,40,41,42} This progress stems from recognises that, while many vital biochemical pathways are universal among plants, there is significant variation in less critical pathways. This variability is increasingly employed in plant classification, highlighting the integration of evidence from multiple sources.

The presence and concentration of phytochemicals in a plant species vary considerably depending on environmental factors, including geographical location ^{43,44,45}. Such variability has been instrumental in characterising medicinal plants, including *Sphenostylis stenocarpa*. Studies have shown that phytochemical analysis serves a crucial function in detecting and classifying landraces of this crop.^{9,46,47,48,49,50,51} This investigation seeks to validate the presence and utility of phytochemical properties in the seeds of *S. stenocarpa*, particularly within its diverse Nigerian landraces. By identifying and quantifying the magnitude of inherent phytochemicals, this study seeks to establish a foundational understanding of the crop's phytochemical profile.

Materials and Methods

Study Location

The study was conducted in the Department of Genetics and Biotechnology at the University of Calabar, Nigeria.

Plant Materials

Five (5) distinct landraces of AYB were obtained from different agricultural farms in January 2024 for this study. The seeds represented entries from Cross River, Abia, Benue, Ogun, and Niger States of Nigeria. These seeds were obtained and planted at the experimental site

located behind the Biological Science block, University of Calabar, Cross River State, Nigeria.

Planting and Cultivation of African Yam Bean

The selected plot was cleared, ploughed, and harrowed to prepare the soil for planting. Miniature mounds were then created with a spacing of 1 metre by 1 metre. Two seeds from each landrace were sown at a depth of 2 cm per mound. Two weeks post-planting, thinning was performed, leaving one plant per mound, which was then staked for support. Weed control was carried out manually as necessary throughout the growth period. Six months after planting, upon reaching maturity, the pods were harvested, and the seeds were extracted for subsequent phytochemical analysis.

Sample collection and preparation of African Yam Bean Extracts

A total of 2 kg of AYB seeds per landrace, obtained using an electronic laboratory weighing balance (Mettler Toledo XP6100), were manually selected and sorted to eliminate stones, damaged seeds, and other foreign materials. The seed samples of *Sphenostylis stenocarpa* were oven-dried at 50°C and subsequently processed into a fine powder using an automated electric blender (Model MS-223, China). The resulting powder was utilized for subsequent analyses. A quantity of 10 g of the *Sphenostylis stenocarpa* seed powder measured using an electronic laboratory weighing balance (*Ohaus* Explorer EX423) was mixed with 100 mL of distilled water at ambient temperature in a conical flask, which was sealed with cotton wool. The mixture was set aside for 24 hours before being sequentially separated using a filtration process through cheesecloth and Whatman No. 1 filter paper. The obtained filtrate was condensed with the help of a rotary evaporator followed by freeze-drying.

Preliminary Phytochemical Screening

A stock solution of crude extract from the AYB was dissolved in 100 mL of ethanol to prepare a solution with 1 g of the extract. This prepared solution was utilised for the phytochemical screening, following the procedure outlined by Nwankwo and Ekeanyanwu, (2023).⁵².

A qualitative analysis of the phytochemicals

The qualitative assessment of the phytochemicals in the aqueous extracts of *Sphenostylis stenocarpa* was performed using established protocols to identify bioactive constituents. The procedures followed standard methodologies.^{53,54,55}

Alkaloid Test

To test for alkaloids, 1 mL of the extract was mixed with 5 mL of 1% aqueous HCl, heated in a steam bath, and then filtered while it was still warm. The residue was dissolved in distilled water, then, 1 mL of the filtrate was treated with a few drops of Mayer's reagent (potassium mercuric iodide solution), Wagner's reagent (iodine and potassium iodide solution), or Dragendorff's reagent (potassium bismuth iodide solution). Alkaloids were identified by the appearance of a cream color with Mayer's reagents.

Tannin Test

For the tannin analysis, 1 mL of the extract was heated in 20 mL of water, followed by filtration. A few drops of 0.1% ferric chloride solution were added to the filtrate. The appearance of a green or blueblack colouration confirmed the presence of tannins.

Test for Flavonoids

Three millilitres of 1% aluminum chloride solution were added to 5 mL of the extract. The development of a yellow colouration indicated the presence of flavonoids. Subsequently, 5 mL of dilute ammonia solution was added to the mixture, followed by concentrated sulphuric acid. The disappearance of the yellow colouration upon standing further confirmed a positive test for flavonoids.

Test for Saponins

Approximately 5 mL of the extract was heated in 20 mL of distilled water using a water bath and subsequently filtered. Ten milliliters of the resulting filtrate were combined with 5 mL of distilled water and shaken vigorously to observe the formation of a stable, persistent froth. To this froth, 3 drops of olive oil were added, and the mixture was shaken again. The appearance of an emulsion confirmed the presence of saponins.

Tannin Test

For the tannin test, 1 mL of the extract was boiled in 20 mL of water, followed by filtration. To the filtrate, a few drops of 0.1% ferric chloride solution were added. The formation of a green or blue-black colouration indicated the presence of tannins.

Test for Terpenoids (Salkowski Test)

To test for terpenoids, 5 mL of the extract was mixed with 2 mL of chloroform and 3 mL of concentrated sulfuric acid was gently added to create a separate layer. The presence of terpenoids was confirmed by the development of a reddish-brown colouration at the interface.

Quantitative assessment of phytochemicals

The phytochemicals found in the aqueous extracts of *Spenostylis* stenocarpa were identified and measured using established methods.

Quantification of Total Alkaloids

A 5 g sample was transferred into a 250 mL beaker, followed by the addition of 200 mL of 10% acetic acid solution prepared in ethanol. The mixture was sealed and left to macerate for 4 hours. It was then filtered, and the resulting filtrate was concentrated using a water bath until the volume was reduced to one-quarter of its initial amount. A concentrated solution of ammonium hydroxide was introduced gradually to the concentrated extract until the precipitation process was fully complete. The mixture was left undisturbed to allow settling of the precipitate, which was subsequently obtained. Dilute ammonium hydroxide was added, and the mixture was then filtered. The residue obtained, indicating the alkaloid content, was dried and weighed.⁵³

Determination of Total Flavonoids

The quantification of total flavonoids was conducted on the development of a flavonoid-aluminum complex, which exhibits a maximum absorbance at 415 nm. A volume of 100 μ L of the sample extract in methanol (10 mg/mL) was combined with 100 μ L of 20% aluminum chloride in methanol and a drop of acetic acid. The mixture was diluted to 5 mL using methanol. After 40 minutes, the absorbance of the solution was measured at 415 nm. Blank samples were prepared by mixing 100 μ L of the extract with a drop of acetic acid and diluting to 5 mL with methanol. A standard solution of rutin (0.5 mg/mL) in methanol was analyzed under identical conditions. All measurements were performed in triplicate to ensure accuracy.⁵⁶

Quantification of Overall Saponins

To determine the saponin content, 20 g of each sample was finely ground and placed in a conical flask. To this, a 100 mL solution of 20% aqueous ethanol was introduced to the sample, and the mixture was then heated in a water bath. The mixture was maintained at 55°C for 4 hours with constant stirring. Once the extraction was done, the mixture was filtered, and the residue was subjected to a second extraction with an additional 200 mL of 20% ethanol. The two filtrates were combined and evaporated to 40 mL in a water bath maintained at around 90°C. The concentrated solution was then moved to a 250 mL separatory funnel, where 20 mL of diethyl ether was introduced. The mixture was agitated thoroughly, and the aqueous phase was separated and retained, while the ether phase was discarded. This purification step was repeated. Following this, 60 mL of n-butanol was added to the aqueous extract, and the combined n-butanol fractions were washed twice with 10 mL of 5% sodium chloride solution. The final solution was evaporated in a water bath and subsequently, it was dried in an oven until a constant

weight was achieved. The amount of saponins present in the sample was analysed based on the weight of the dried residue.⁵⁷

Quantification of overall Tannins

An accurately weighed 500 mg sample was placed into a 50 mL plastic bottle. To this, fifty millilitres of distilled water were added, and the mixture was agitated for 1 hour using a mechanical shaker. Afterward, the filtrate was collected in a 50 mL volumetric flask, which was then filled to the mark with distilled water. A 5 mL aliquot of the filtrate was transferred into a test tube, followed by the addition of 2 mL of 0.1 M FeCl₃ in 0.1 N HCl and 0.008 M potassium ferrocyanide were added. The absorbance of the resulting solution was then measured at 120 nm within 10 minutes.⁵⁸

Determination of Terpenoids

A 100 mg portion of dried *Sphenostylis stenocarpa* extract (denoted as *wi*) was placed in 9 mL of ethanol and allowed to soak for 24 hours. After filtration, the extract was subjected to a second extraction with 10 mL of petroleum ether using a separating funnel. The ether phase was collected into pre-weighed glass vials and allowed to dry completely. Once the ether had evaporated, the final weight of the dried residue (*wf*) was recorded. The total terpenoid content was calculated as a percentage using the following equation: (Wi-wF/ wi x 100) where *Wi* represents the initial weight of the sample and *WF* is the weight after evaporation of the ether.⁵⁹

Data Analysis

The qualitative data were summarized using descriptive statistics, as outlined by Nwanko & Ganyam (2021)⁵¹. For the quantitative data, analysis of variance (ANOVA) was performed, and mean differences were separated using the Least Significant Difference (LSD) test. Predictive Analytics Software (PASW) version 20.0 was used.

Results And Discussion

Qualitative analysis

The substantial genotypic variation observed in the phytochemical content of African yam bean (*Sphenostylis stenocarpa*) seeds highlights the species' strong genetic potential for advancing targeted crop improvement and sustainable breeding initiatives. Phytochemical analyses of five distinct landraces from various regions revealed notable diversity in their phytochemical profiles. A detailed qualitative assessment further confirmed significant variability in the phytochemical composition among the studied landraces. Several studies have confirmed the presence of antinutritional factors in African yam bean seeds.^{60,61,62,63,64}

For the qualitative analysis (Table 1), the legend; + (detected), - (absent) was used to qualify the amount of phytochemical in each of the selected landraces of AYB. Following the qualitative analysis through various specific tests, different phytochemicals were discovered in good amounts across each of the selected landraces respectively.

Alkaloids were consistently detected in moderate concentrations across all examined landraces, a finding consistent with earlier reports on *Sphenostylis stenocarpa*, though variations in alkaloid levels have been noted ⁹. This observation suggests that alkaloid production is a stable trait within the African yam bean (AYB) species. The consistency across landraces indicates that the genetic or environmental factors governing alkaloid synthesis are relatively uniform among the studied accessions.

Flavonoids, on the other hand, were highly detected in the Benue landraces of AYB, moderately present in the landraces from Cross River, Abia, and Ogun, and only slightly detected in the Niger State landraces. Flavonoid abundance in the Benue landraces is consistent with findings by Haytowitz *et al.*, (2013) ⁶⁵ which demonstrated that the distribution of bioactive compounds, such as flavonoids, varies across different accessions. The elevated flavonoid levels in the Benue landraces may point to region-specific genetic or environmental factors that influence flavonoid biosynthesis.⁶⁶ In contrast, other landraces exhibited varying flavonoid concentrations, underscoring the impact of regional variability.

High levels of saponins were observed in the Cross River and Abia landraces, while moderate concentrations were detected in the Niger landrace, and lower levels were found in the Benue and Ogun landraces. This variability aligns with the findings of Popoola *et al.*, $(2023)^3$ who reported differential saponin concentrations across various *S. stenocarpa* landraces. The moderate saponin levels in the Niger

landrace corroborate the work of Nwankwo and Ganyam, (2021)⁵¹ who noted similar moderate saponin concentrations in African yam bean milk extracts. The elevated saponin content in the Cross River and Abia landraces may suggest the influence of specific ecological or genetic factors driving saponin production in these regions.

Specific Test	Phytochemical Compounds	Cross River	Abia	Benue	Ogun	Niger
Wagner	Alkaloids	+	+	+	+	+
Lead Acetate	Flavonoids	+	+	+	+	+
Froth	Saponins	+	+	+	+	+
Ferric chloride	Tannins	+	+	+	+	+
Salkowski	Terpenoids	+	+	+	+	+

Legend: + = detected, - = absence.

Tannins were consistently present in all landraces, with the Benue landrace exhibiting the highest levels. This high tannin presence is in agreement with the findings of Shitta *et al.*, (2022)⁶⁷ who also reported elevated tannin content in *S. stenocarpa*. The widespread presence of tannins could indicate a core characteristic of the species or an adaptation to particular ecological conditions.

Terpenoids were detected in relatively low amounts across most landraces, with the Niger landrace showing moderate levels. In contrast to our findings, Popoola *et al.*, (2023) ³ reported that legumes, including underutilized species, often contain terpenoids, among other bioactive compounds.

Quantitative analysis

Significant variation in quantitative traits within a species is a fundamental concept in biology. This research highlights notable differences in the phytochemical profiles of various African yam bean (AYB) accessions, aligning with previous studies that have consistently reported diversity across multiple aspects of AYB.^{60,61,62,63,68}

The quantitative analysis of five phytochemicals across African yam bean (AYB) landraces is summarised in Table 2, with a p-value > 0.05. The ANOVA results in Table 3 confirmed that there were no statistically significant differences (P > 0.05) in the concentrations of alkaloids, flavonoids, saponins, tannins, and terpenoids among the five landraces. While the mean concentrations of these phytochemicals varied across the landraces, the observed differences were not significant, highlighting a level of consistency in their distribution across the sampled populations. Leguminous seeds are recognised for their substantial antioxidant content, attributed to their elevated levels of phenolics, flavonoids, and anthocyanins. Products derived from these legumes have the potential to contribute to the prevention and management of chronic and degenerative diseases while addressing issues of protein-energy malnutrition.^{69,70} According to Soetan *et al.*, (2018)27 the antioxidant levels among different African yam bean accessions demonstrated significant variation. Notably, our findings reveal a remarkable consistency in phytochemical concentrations across the sampled regions, presenting a contrast to the qualitative variability reported in previous studies.

Table 2:	Quantitative	analysis o	of five phy	<i>tochemicals</i>	in fiv	e AYB	landraces
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Alkaloids (mg/100g)	Flavonoids (mg/100g)	Saponins (mg/100g)	Tannins (mg/100g)	Terpenoids (mg/100g)
28.00 ^a ±0.577	7.00 ^a ±0.577	3.33 ^a ±0.882	2.500 ^a ±0.2887	5.00 ^a ±0.577
25.67 ^a ±2.963	5.33 ^a ±0.882	3.67 ^a ±0.667	1.833 ^a ±0.4410	4.33 ^a ±0.882
27.33 ^a ±4.256	7.67 ^a ±0.882	4.67 ^a ±0.333	$3.000^{a}\pm0.0000$	6.33 ^a ±0.333
25.00 ^a ±2.309	7.00 ^a ±1.000	3.67 ^a ±0.333	2.000 ^a ±1.0000	5.33 ^a ±1.667
26.33 ^a ±2.028	$7.00^{a} \pm .577$	$3.67^{a}\pm0.667$	2.333 ^a ±0.3333	$5.00^{a}\pm1.155$
	Alkaloids (mg/100g) 28.00 ^a ±0.577 25.67 ^a ±2.963 27.33 ^a ±4.256 25.00 ^a ±2.309 26.33 ^a ±2.028	Alkaloids Flavonoids (mg/100g) (mg/100g) $28.00^{a}\pm 0.577$ $7.00^{a}\pm 0.577$ $25.67^{a}\pm 2.963$ $5.33^{a}\pm 0.882$ $27.33^{a}\pm 4.256$ $7.67^{a}\pm 0.882$ $25.00^{a}\pm 2.309$ $7.00^{a}\pm 1.000$ $26.33^{a}\pm 2.028$ $7.00^{a}\pm .577$	Alkaloids Flavonoids Saponins (mg/100g) (mg/100g) (mg/100g) $28.00^{a}\pm 0.577$ $7.00^{a}\pm 0.577$ $3.33^{a}\pm 0.882$ $25.67^{a}\pm 2.963$ $5.33^{a}\pm 0.882$ $3.67^{a}\pm 0.667$ $27.33^{a}\pm 4.256$ $7.67^{a}\pm 0.882$ $4.67^{a}\pm 0.333$ $25.00^{a}\pm 2.309$ $7.00^{a}\pm 1.000$ $3.67^{a}\pm 0.667$ $26.33^{a}\pm 2.028$ $7.00^{a}\pm .577$ $3.67^{a}\pm 0.667$	AlkaloidsFlavonoidsSaponinsTannins(mg/100g)(mg/100g)(mg/100g)(mg/100g) $28.00^{a}\pm0.577$ $7.00^{a}\pm0.577$ $3.33^{a}\pm0.882$ $2.500^{a}\pm0.2887$ $25.67^{a}\pm2.963$ $5.33^{a}\pm0.882$ $3.67^{a}\pm0.667$ $1.833^{a}\pm0.4410$ $27.33^{a}\pm4.256$ $7.67^{a}\pm0.882$ $4.67^{a}\pm0.333$ $3.000^{a}\pm0.0000$ $25.00^{a}\pm2.309$ $7.00^{a}\pm1.000$ $3.67^{a}\pm0.333$ $2.000^{a}\pm1.0000$ $26.33^{a}\pm2.028$ $7.00^{a}\pm.577$ $3.67^{a}\pm0.667$ $2.333^{a}\pm0.3333$

*Means expressed by the same superscript case letter along the vertical axis indicate no significant difference.

Table 3: A	nalvsis of	variance f	for five p	hvtoche	emicals i	n African	vam beans	obtained from	five states	in Nigeria
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		Sum of Squares	df	Mean Square	F	Sig.
	Between Groups	17.733	4	4.433	0.202	0.932
Alkaloids	Within Groups	220.000	10	22.000		
	Total	237.733	14			
	Between Groups	9.067	4	2.267	1.172	1.380
flavonoids	Within Groups	19.333	10	1.933		
	Total	28.400	14			
	Between Groups	3.067	4	0.767	0.676	0.684
Saponins	Within Groups	11.333	10	1.133		
	Total	14.400	14			
	Between Groups	2.500	4	0.625	0.750	0.780
Tannins	Within Groups	8.333	10	0.833		
	Total	10.833	14			
	Between Groups	6.400	4	1.600	0.500	0.737
Terpenoids	Within Groups	32.000	10	3.200		
-	Total	38.400	14			

The analysis of alkaloid content among the five (5) African yam bean (AYB) landraces showed that the Cross River collection had the highest mean concentration (28.00 mg/g), followed by Benue (27.33 mg/g) and Niger State (26.33 mg/g), while the Ogun collection recorded the lowest concentration (25.00 mg/g). These findings align with the observations of Anya and Ozung, $(2019)^{71}$ who reported elevated alkaloid levels in AYB seeds. The high alkaloid levels, particularly of rotenone, present nutritional concerns and pose a significant limitation to the wider utilisation of this crop.⁶⁵ Nevertheless, no significant difference was observed (P>0.05) in alkaloid content among the landraces, suggests that genetic variability in alkaloid production might be limited. Alternatively, environmental influences could play a more significant role, potentially overshadowing genetic factors in determining alkaloid concentrations.

The flavonoid content analysis of the five (5) African yam bean (AYB) collections showed no significant differences (P>0.05) among the landraces. The Benue State collection had the highest flavonoid concentration (7.67 mg/g), followed by Cross River, Ogun, and Niger State, each with a mean of 7.00 mg/g, while Abia State recorded the lowest value (5.33 mg/g). These findings indicate a general consistency in flavonoid levels across the landraces, aligning with previous research^{71,72,73,74} that identifies various factors, including cultivar type, as determinants of flavonoid variability. The elevated flavonoid levels observed in the Benue collection may warrant further exploration, particularly if enhancing flavonoid content is a goal for breeding programmes or nutritional fortification initiatives.

The saponin content analysis indicated that the Benue collection had the highest concentration (4.67 mg/g), followed by Abia, Ogun, and Niger, each with 3.67 mg/g, while Cross River recorded the lowest saponin content (3.33 mg/g). Similarly, the tannin content analysis revealed no significant differences (P>0.05) among the landraces. However, Benue recorded the highest tannin concentration (3.00 mg/g), followed by Cross River (2.50 mg/g) and Ogun (2.00 mg/g), with Abia State having the lowest tannin content (1.83 mg/g). Despite these variations, the insignificant differences suggest that the levels of saponins and tannins are generally consistent across the landraces. The observed differences are unlikely to result from distinct genetic or environmental factors, reflecting a pattern consistent with findings by Ajibola and Olapade, (2016) ⁷⁵ who reported no significant variation in saponin and tannin concentrations across five accessions of Sphenostylis stenocarpa. Nevertheless, the relatively higher concentrations observed in the Benue collection, particularly for tannins, and the lower values in the Abia collection may indicate the influence of subtle regional or environmental factors or minor genetic variability.

Benue recorded the highest terpenoid content (6.33 mg/g), followed by Ogun (5.33 mg/g), with both Cross River and Niger showing 5.00 mg/g. The Abia State collection exhibited the lowest terpenoid concentration (4.33 mg/g). Insignificant differences (P>0.05) in terpenoid levels among the African yam bean (AYB) landraces suggest that terpenoid production may be less affected by geographic variability compared to other phytochemicals. This consistency implies that factors other than geographic location, such as genetic factors or environmental influences, may have a minimal impact on terpenoid synthesis across the landraces studied.

Among the landraces, the Benue collection stands out as the most suitable candidate for conservation efforts, given its consistently high concentrations of multiple phytochemicals. Furthermore, the Cross River landrace is valuable for preservation due to its alkaloid-rich profile, which contributes to maintaining both genetic and biochemical diversity within African yam bean (AYB) populations. These collections could play a crucial role in ensuring the sustainable use and enhancement of AYB's nutritional and agronomic potential.

Conclusion

This study offers an in-depth analysis of the phytochemical composition across five landraces of AYB from different states in Nigeria. Qualitative analysis revealed notable regional variability in the presence of alkaloids, flavonoids, saponins, tannins, and terpenoids across the landraces. However, quantitative analysis revealed no significant differences (P>0.05) in the concentrations of these phytochemicals, indicating a general consistency in their levels despite qualitative differences.

The Benue landrace, which exhibited the highest flavonoid content, emerges as a strong candidate for its potential nutritional and medicinal benefits. However, its relatively low saponin content may limit its use for specific applications where saponins are prioritised. On the other hand, the Cross River and Abia landraces, with high saponin and tannin levels, represent valuable resources for conservation, especially if these secondary metabolites are of particular interest for future research or crop improvement programs. The Niger landrace, with its unique moderate terpenoid content, also presents an intriguing opportunity for specialized applications in biotechnology or agriculture.

In terms of conservation, the Benue collection is identified as the most suitable due to its consistent phytochemical profile, while the Cross River landrace, known for its alkaloid-rich composition, holds significant value for maintaining genetic and biochemical diversity within AYB populations. Together, these collections are essential for ensuring the sustainable use and enhancement of the nutritional and agronomic potential of African yam beans.

Conflict of Interest

The authors have no conflicts of interest to disclose.

Authors' Declaration

The authors affirm that the research presented in this article is original and take full responsibility for any claims associated with its content.

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