







## Original Article

## Phytochemical screening and evaluation of antioxidant properties of African locust bean marketed in Ibadan, Nigeria



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### ABSTRACT

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This study evaluated the phytochemical and antioxidant properties of African locust beans marketed in Ibadan metropolis. The nutritional qualities of food consumed should be of great importance to consumers and the populace at large. Hence, there is a need for investigation into African locust beans sold in the Ibadan metropolis. Three markets each were investigated within the three local governments, and they are: Muslim, Olomi, and Orita Challenge in Ibadan South East local government, Agbon, Academy, and Monatan in Lagelu local government, and Eleyele, Dugbe, and Oje in Ibadan North West local government. Fresh locust bean samples were obtained from these markets, labelled, and transported to the laboratory for analysis. Standard procedures were carried out to determine the Antioxidant potentials and Phytochemical screening of processed *Parkia biglobosa* sourced from the various markets. The result of the qualitative phytochemical screening revealed the presence of some bioactive compounds, which are Saponins, Terpenoids, Alkaloids, Flavonoids, Cardiac glycosides, and Anthraquinones, while Tanins, Steroids, and Phenols are generally absent in the three local governments under investigation. The percentage scavenging of locust bean samples from Lagelu was the highest, ranging from 52.68 to 63.77%, followed by samples from Ibadan South East, with values ranging from 29.92 to 58.57%. The least scavenging percentage was observed in Ibadan North West, ranging from 30.50 to 37.48. The presence of these bioactive compounds and secondary metabolites suggests the potential ability of African locust beans to improve human health if included in diets.

### INTRODUCTION

Fermented locust bean seed, also known locally as "dawadawa" or "iru" in Nigeria (Yoruba), plays a significant role in culinary tradition and offers various health benefits (Zebedee *et al.*, 2022). Fermented locust beans are commonly used as a flavor

enhancer and seasoning in traditional African cuisines (Adeoye, 2018). The fermentation process develops a rich, savory, and pleasant taste, which adds depth and complexity to dishes. Fermented locust beans contribute to the unique taste and aroma of African soups, stews, sauces, and condiments (Akubor *et al.*, 2017). Their incorporation in cooking brings diversity to flavor

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and contributes to the overall sensory appeal of cuisine (Akubor *et al.*, 2017; Ezugwu *et al.*, 2018). The use of condiments in food does not stand for nutrition alone, but is medicinal in nature based on their contribution to the overall health of humans. The use of herbs or plant products by man as medicine has been from time immemorial. The World Health Organization (WHO) reports that more than 80 % of the world's population relies on traditional medicine for their primary health care needs, especially in developing countries (Awuchi *et al.*, 2021). An estimated 25 % of modern medicines are made from plants, first used as traditional medicine in the treatment of diseases (De Vries and Visser, 2021). In rural areas, there are additional cultural factors that encourage the use of herbal preparations; people believe that where a particular disease is endemic, there shall always be plants to cure such an ailment within the environment. Other factors are the absence of primary health care centers, lack of competent staff, poor diagnostic facilities, and inadequate supplies of medicines (Daramola, 2014).

Owing to the development of adverse effects and microbial resistance to chemically synthetic drugs, there is a global shift to ethno-pharmacognosy. Many health-beneficial activities of plants, such as anticancer, antimicrobial, antioxidant, antidiarrheal, analgesic, and wound healing activity, were reported as a result of studies conducted on medicinal plants and their seeds (Eboma *et al.*, 2020). Recently, medicinal plants have become the focus of intense study in terms of conservation and their traditional medicinal uses. The studies on plants have either supported the claims by the traditional healers or contradicted their folkloric beliefs (Esenwah and Ikenebomeh, 2018).

In recent years, there has been a growing interest in the utilization of traditional African foods for their potential health benefits. African locust beans, derived from the tree *Parkia biglobosa*, are a staple in many West African cuisines, valued for their unique flavor and nutritional properties. Beyond their culinary significance, African locust beans have been traditionally believed to possess medicinal qualities due to their rich phytochemical composition. In Ibadan Metropolis, Nigeria, African locust beans hold particular cultural and economic importance, being widely consumed and traded. However, despite its widespread use, there remains a gap in scientific understanding regarding the availability of different phytochemical compounds and abundant antioxidant properties in African locust beans sold in the Ibadan metropolis. Currently, the demands remain consistently high with consumers showing a strong preference for this culinary ingredient (Ijewere, 2019). Therefore, this study aims to examine the phytochemicals and determine the antioxidant capacity of African locust beans sourced from various markets across Ibadan Metropolis.

## MATERIALS AND METHODS

### Description of study area

Commercially available locust beans were purchased from markets in each of the three local government areas in Ibadan, which are Muslim, Olomi, and Orita challenge in Ibadan South East Local Government Area; Agbon, Academy, and Monatan in Lagelu Local Government Area; and Eleyele, Dugbe, and Oje in Ibadan North West Local Government Area. The locust bean samples were analyzed for antioxidant and phytochemical properties. The local government areas are shown in Figure 1.

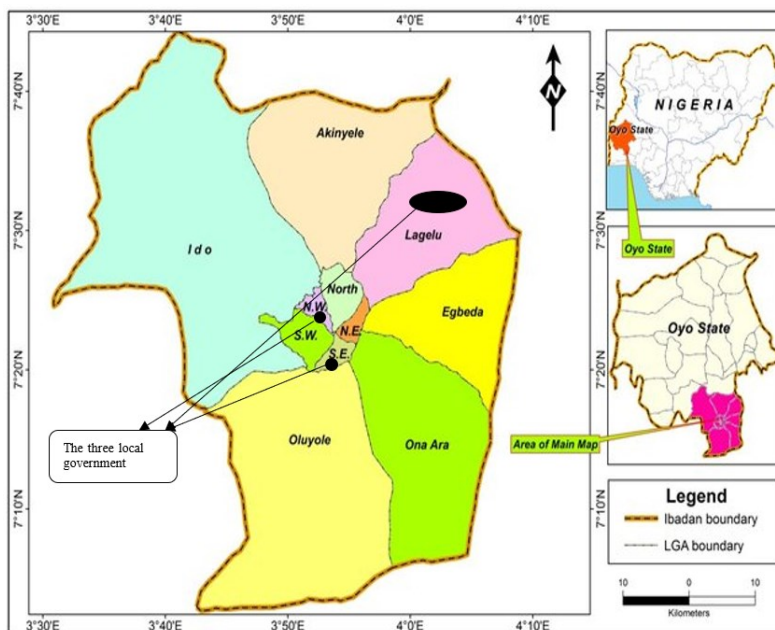


Figure 1: Map showing the local government areas of Oyo State, including Lagelu, Ibadan North West, and Ibadan South East local governments.



## Sample preparation

African locust bean was oven dried at 50°C for 48 hours to obtain a moisture content of 12%. Samples of the African locust beans were ground and sieved with a wire mesh of 0.1mm to ensure a uniform particle size. Samples were stored in glass jars to prevent absorption of moisture and contamination. The prepared samples were mixed with ethanol in a ratio of 1:10 and left for 24 hours. The filtrates of the mixture were evaporated to obtain the crude extract.

## Qualitative Phytochemical Screening Analysis

Qualitative phytochemical screening of the locust bean samples was carried out using the standard procedures as described by Sofowora (2008). Bioactive compounds, which include Tannins, Saponins, Flavonoids, Terpenoids, Glycosides, Alkaloids, Anthraquinones, Steroids, and

Phenol was determined using standard procedures.

## Evaluation of Antioxidant Properties

### DPPH scavenging activity

In order to evaluate the antioxidant potential through free radical scavenging by the test samples, the change in optical density of DPPH radicals was monitored. According to Manzocco *et al.* (2018).

### Ferric ion Reducing Antioxidant Power assay (FRAP)

The locust bean samples in different concentrations were mixed with 2.5ml of 20 mM phosphate buffer and 2.5 mL 1%, w/v potassium ferricyanide, and then the mixture was incubated at 50 °C for 30min. Afterwards, 2.5 ml of 10%, w/v trichloroacetic acid and 0.5ml 0.1%, w/v ferric chloride were added to the mixture, which was kept aside for 10 min. Finally, the absorbance was measured at 700 nm. Ascorbic acid was used as a positive reference standard. All assays were run in triplicate, weighed, and averaged (Manzocco *et al.*, 2018).

## RESULTS AND DISCUSSION

### Results

There is an abundance of Saponins and Terpenoids in all samples of all three markets. Flavonoids are abundant in Muslim market samples. Cardiac glycosides and Anthraquinones are present in sizeable amounts across the three markets. Olomi has an abundance of Alkaloids. While Steroids and phenols are absent. The locust beans marketed in Ibadan South East local government have an abundance of Saponins and Terpenoids, a regular amount of Glycosides and Anthraquinones, and varied amounts of flavonoids and alkaloids. There is an absence of steroids and phenols in random samples across the local government (Table 1).

Saponins and Terpenoids are abundant in all samples, while Alkaloids are abundant in Agbon and Academy samples. Flavonoids, Cardiac glycosides, and Anthraquinones are present in all markets. While Tannins, Steroids, and Phenols are generally absent in the samples, according to the local government. (Table 2).

Saponin is abundant in all three markets. Terpenoids are abundant in Eleyele and Dugbe markets, but are also present in the Oje market. Eleyele and Dugbe market samples show an abundance of alkaloids, but they are moderately present in the Oje market. Flavonoids, Cardiac glycosides, and Anthraquinones are present in all three markets, while they show the absence of phenols, tannins, and steroids. The result of the phytochemical screening shows that locust beans obtained across the three study local government areas in Ibadan generally have an abundance of Saponins and Terpenoids with above moderate amounts of Flavonoids and Alkaloids, while Cardiac glycosides and Anthraquinones are present in normal amounts with a generally non-consistent consistency of Tannins, Steroids, and Phenols (Table 3).

**Table 1: Table showing the results of qualitative phytochemical screening for the different locust bean samples collected from Ibadan South East Local Government**

Test	L1M1S1	L1M1S2	L1M1S3	L1M2S1	L1M2S1	L1M2S2	L1M3S1	L1M3S2	L1M3S3
Saponins	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve
Tannins	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Flavonoids	++ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Cardiac glycosides	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Anthraquinones	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Terpenoids	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve
Steroids	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Alkaloids	++ve	+ve	+ve	++ve	++ve	++ve	+ve	+ve	+ve
Phenol	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve

Key: the intensity of abundance of phytochemicals is shown as ++ve: Abundant, +ve: Present, -ve: Absent, L1: Ibadan South East Local Government, M1: Muslim Market, M2: Olomi Market, M3: Orita Challenge Market, S1, S2, S3: Sample 1, Sample 2, and Sample 3, respectively.



**Table 2: Table showing the results of qualitative phytochemical screening for the different locust bean samples collected from Lagelu local Government**

Test	L2M1S1	L2M1S2	L2M1S3	L2M2S1	L2M2S2	L2M2S2	L2M3S1	L2M3S2	L2M3S3
Saponins	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve
Tannins	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Flavonoids	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
cardiac glycosides	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Anthraquinones	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Terpenoids	++ve	++ve	++ve	++ve	++ve	++ve	++ve	+ve	++ve
Steroids	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Alkaloids	++ve	++ve	+ve	++ve	++ve	++ve	+ve	+ve	+ve
Phenol	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve

Key: the intensity of abundance of phytochemicals is shown as L2: Lagelu Local Government, M1: Agbon Market, M2: Academy Market, M3: Monatan Market, S1, S2, S3: Sample 1, Sample 2, and Sample 3, respectively.

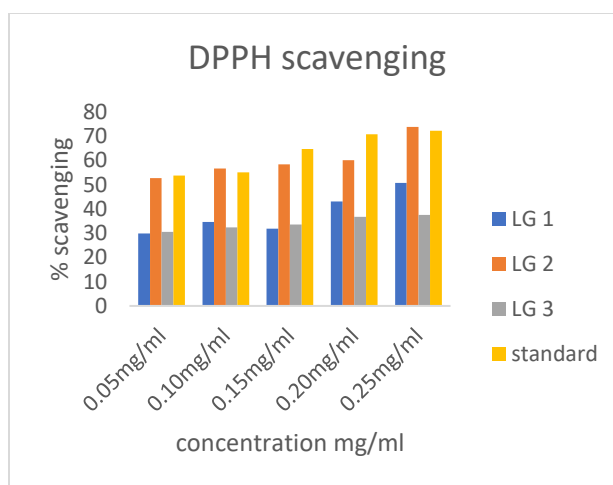
**Table 3: Table showing the results of qualitative phytochemical screening for the different locust bean samples collected from Ibadan North West Local Government**

Test	L3M1S1	L3M1S2	L3M1S3	L3M2S1	L3M2S2	L3M2S2	L3M3S1	L3M3S2	L3M3S3
Saponins	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve	++ve
Tannins	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Flavonoids	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Cardiac glycosides	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Anthraquinones	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve	+ve
Terpenoids	++ve	++ve	++ve	++ve	++ve	++ve	+ve	+ve	+ve
Steroids	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve
Alkaloids	++ve	++ve	++ve	++ve	++ve	++ve	+ve	+ve	++ve
Phenol	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve	-ve

Key: L3: Ibadan North West Local Government, M1: Eleyele Market, M2: Dugbe Market, M3: Oje Market. S1, S2, S3: Sample 1, Sample 2 and Sample 3 respectively.

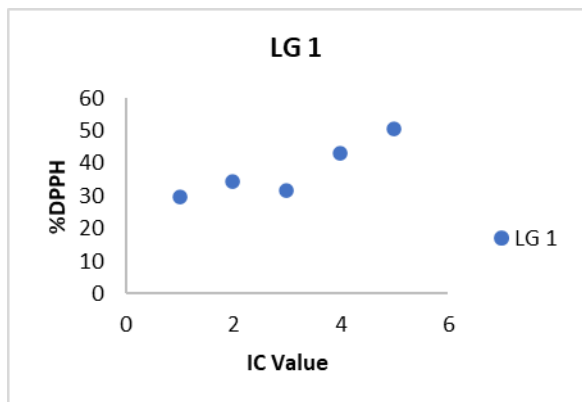
### Antioxidant properties using the DPPH scavenging parameter

The vertical axis of the graph represents the percentage scavenging by the locust bean samples, while the horizontal axis represents the concentrations of locust bean samples used. The results of the antioxidant with DPPH scavenging parameters, as shown in Figure 2, reveal that the sample collected from Lagelu local government scavenged free radicals through the different concentrations of the samples increased across the three local government areas, the scavenging properties also increased. Lagelu showed  $52.68 \pm 0.24$  scavenging activity at the lowest concentration used and  $63.77 \pm 0.06$  scavenging activity at the highest concentration used. This is higher than Ibadan South East,  $29.92 \pm 0.17$  to  $50.67 \pm 0.2$ , and Ibadan North West,  $30.50 \pm 0.33$  to  $37.48 \pm 0.19$ . But lower than that of the standard (ascorbic acid) with  $53.80 \pm 0.34$  at the lowest concentration and  $72.24 \pm 0.66$  at the highest concentration (Figure 1).

**Figure 2: Histogram representation of the % scavenging of the different samples against the standard**

**Calculated IC<sub>50</sub> Values**

The IC<sub>50</sub> is the concentration of locust bean sample needed to inhibit the free radicals (scavenge) by 50%. It is called the half maximal inhibitory concentration. Usually, the antioxidant properties of a plant or any other food substance are measured by the ability of its components to neutralize free radicals by reducing them (Figure 2).

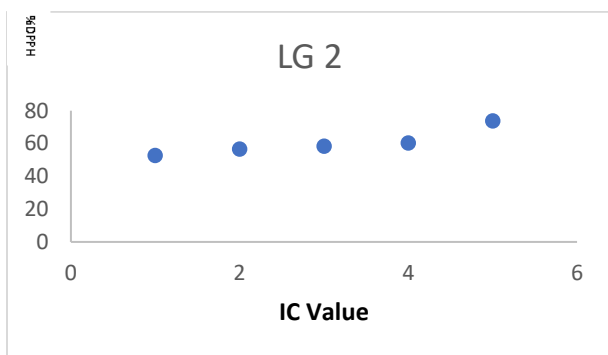


**Figure 3: Calculated IC<sub>50</sub> for the sample collected from Ibadan South East Local Government (LG 1)**

The y axis represents the % free radical inhibited (that is, the % DPPH scavenged) while the x axis represents the IC value. The IC<sub>50</sub> value is the IC value that corresponds to 50 on the vertical axis. From the graph equation,  $y = mx + c$ , where y is the value on the y-axis, m is the slope of the graph, x is the x-axis value, and c is the intercept, the IC<sub>50</sub> value is x (amount of locust beans) when y (% free radicals inhibited) is 50. Slope (m) = 5.001, intercept (c) = 23.029, at y = 50, IC<sub>50</sub> value is calculated below:

$$50 = 5.001x + 23.029$$

IC<sub>50</sub> for the sample from LG 1 is 5.39

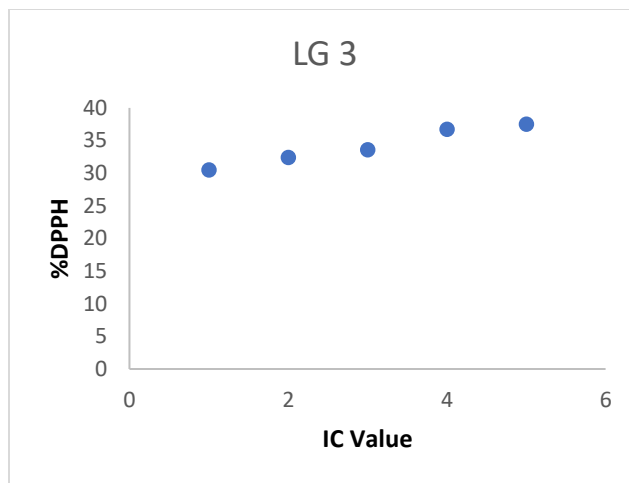


**Figure 4: Calculated IC<sub>50</sub> for the sample collected from the Lagelu local government (LG 2)**

IC<sub>50</sub> for the sample collected from LG 2. Slope (m) = 4.571, Intercept (c) = 46.587, therefore x value at y=50 is calculated as:

$$50 = 4.571x + 46.587$$

IC<sub>50</sub> for the LG 2 sample is 0.75. The IC<sub>50</sub> of the sample collected from Lagelu local government (LG 2) (0.75) was calculated to be the lowest



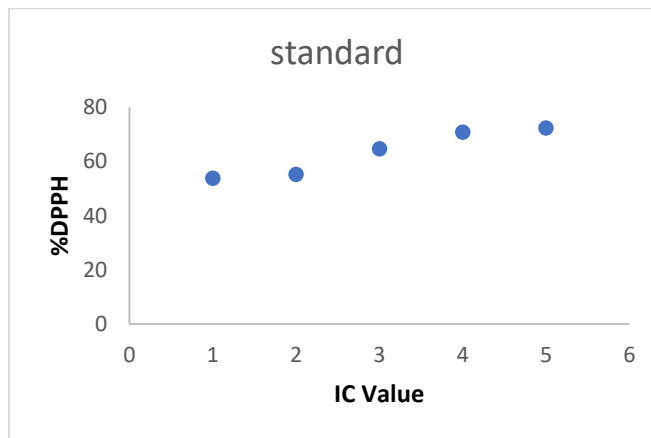
**Figure 5: Calculated IC<sub>50</sub> for the sample collected from Ibadan North West local government (LG 3)**

IC<sub>50</sub> for the sample collected from LG 3

Slope of the graph (m) = 1.826, intercept (c) = 28.654, value of x at y=50 IC<sub>50</sub> is calculated:

$$50 = 1.826x + 28.654$$

IC<sub>50</sub> for the LG 3 sample is 11.69. Calculated IC<sub>50</sub> for the standard (ascorbic acid)



**Figure 6: IC<sub>50</sub> for the standard (ascorbic acid)**

Slope = 5.247, Intercept = 47.565, value of x at y=50 is calculated:

$$50 = 5.247x + 47.565$$

IC<sub>50</sub> for the standard is 0.4641

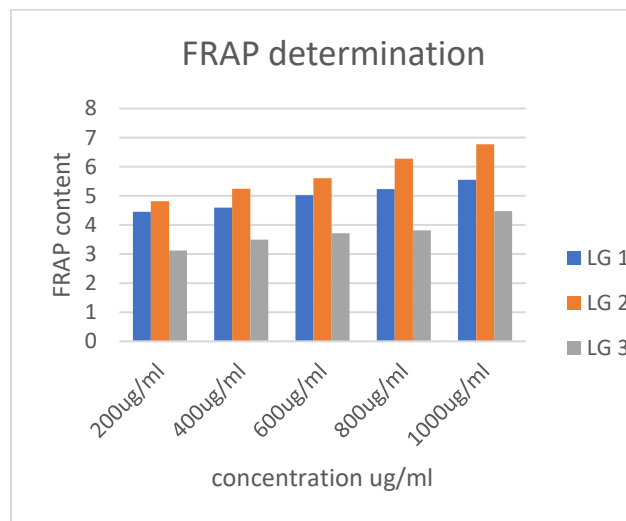


The IC<sub>50</sub> of the standard (0.46) is quite lower than that of the different samples.

### Antioxidant using the FRAP (Ferric Reducing Antioxidant Properties) parameter

The following are the respective calculated FRAP content at ascorbic acid equivalence (AAE) in the different samples:

The results of the antioxidant evaluation using FRAP, as shown in Table 4, showed that the sample from local government area 2 (Lagelu) reduced free radicals compared to samples from other local governments



**Figure 7: Histogram representation of the FRAP content of the different samples at different concentrations of Ascorbic Acid Equivalence (AAE)**

### Discussion

The result of samples from the three local government areas revealed an abundance of phytochemical constituents such as Saponins, Terpenoids, Alkaloids, Glycosides, and Anthraquinones in moderate amounts across the three local government areas, while phenols and steroids are absent. Flavonoids were also present in large amounts in some samples. The result obtained from this study revealed the presence of secondary metabolites, which could reflect the importance of dietary phytochemicals in promoting well-being and preventing chronic diseases, which is widely recognized in the vast field of nutrition science. Some experimental studies support the result obtained in this study, which showed that the range of biological activities associated with these plant-derived compounds demonstrates their potential benefits for human health (Salehi *et al.*, 2020; Eboma *et al.*, 2021; Aremu *et al.*, 2015). Flavonoids are secondary metabolites that play important roles in protecting plants against damage and improving plant aroma, coloration, and flavor. They are potent water-soluble antioxidants and free radical scavengers that prevent oxidative cell damage, have strong anticancer activity,

and protect against the different levels of carcinogenesis (Oluwaniyi & Bazambo, 2014; Oloyede & Akintunde, 2019). African locust bean is rich in phytochemicals such as flavonoids, phenolic compounds, tannins, and alkaloids. These compounds are known to exhibit antioxidant activity by scavenging free radicals and inhibiting oxidative stress in the body (Akintobi *et al.*, 2016). Ezugwu *et al.* (2018) revealed that Tannin is one of the metal chelating compounds that precipitate Proteins. They are also known for their anti-oxidant and anti-microbial properties. Saponins lower the cholesterol level; they have anti-diabetic and anti-carcinogenic properties. Some of the secondary metabolites, like Saponins, Alkaloids, Steroids, and Flavonoids, have been discovered to be starting materials for most of the synthetic drugs. The Alkaloids are specifically used as analgesics, stimulants, anesthetics, and antibacterials. Alkaloids are said to be pharmacologically active, and their actions are felt in the autonomic nervous system, blood vessels, promotion of diuresis, respiratory system, gastrointestinal tract, uterus, and malignant diseases. However, it was also noted that the quantity of the secondary metabolites in the plant samples varies from one location to another. Therefore, further phytochemical screening should be done on the African locust bean seeds to ascertain their curative properties; the phytochemicals found in them have good potential for the development of bioactive drugs and other pharmaceutical products if well harnessed.

Samples from the three local government areas demonstrated different degrees of antioxidant properties, with samples from the Lagelu local government area showing a high antioxidant potential. Samples from other markets (local government areas) also showed considerable antioxidant power, ascertaining the potential of African locust bean in nutritional and health applications. The qualities/characteristics of the African locust beans could be employed for nutrition and health uses/applications, and have promising beneficial applications in health and medicine in the future if the benefits are properly screened, developed, and harnessed for economic and medical uses. The seeds of African locust bean contain vitamins such as vitamin C, vitamin A (as beta-carotene), and vitamin E, all of which are potent antioxidants. These vitamins help neutralize harmful free radicals, reducing oxidative damage to cells and tissues (Ajayi *et al.*, 2016). Free radicals are highly reactive molecules that can damage cells and contribute to various diseases, including cancer and cardiovascular diseases. The result of the antioxidants present in African locust bean supports the findings by Oluwaniyi & Bazambo (2014), which explain the effect of antioxidants in neutralizing these free radicals, thereby protecting cells from oxidative damage.

The antioxidant compounds in African locust bean help prevent lipid peroxidation, maintaining the integrity of cell membranes (Oguntoyinbo & Sanni, 2017). Oxidative stress often goes hand in hand with inflammation. African locust bean has been found to possess anti-inflammatory properties, which can further contribute to its antioxidant effects by reducing inflammation-mediated oxidative damage (Tijani *et al.*, 2019). The antioxidant



properties of African locust bean contribute to its potential health benefits, including its ability to support cardiovascular health, reduce the risk of chronic diseases such as cancer, diabetes, and neurodegenerative disorders, and promote overall well-being (Iyamu *et al.*, 2014). In conclusion, African locust bean is a valuable source of antioxidants due to its rich phytochemical composition and vitamin content. Regular consumption of African locust bean and its products may help protect against oxidative stress-related damage and promote health and longevity. However, further research is needed to explore its full antioxidant potential and its implications for human health.

## CONCLUSION AND RECOMMENDATIONS

In conclusion, the phytochemical properties and antioxidant potential of African locust beans sold in Ibadan metropolis were studied. This study revealed the presence and the level of phytochemicals and antioxidants in the locust beans sold within the Ibadan metropolis. Thus, phytochemical screening revealed the presence of some bioactive compounds, which are Saponins, Terpenoids, Alkaloids, Flavonoids, Cardiac glycosides, and Anthraquinones, while Tanins, Steroids, and Phenols are generally absent in the three local governments under investigation. The study also revealed that the antioxidant properties exhibited in these samples increase as the concentration increases in comparison with the ascorbic standard. The results obtained from this study could be used as preliminary data for further research studies on African locust beans in relation to their use as biomedicine. However, it is recommended from this study that African Locust beans should be included as a condiment in household meals to boost the bioactive compounds present in our meals.

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## Authors contributions

Author AIT collated and interpreted data and wrote the manuscript. AEA proofread and edited the manuscript. FAK collected and prepared the samples for analysis and wrote the introduction. OEM and ORI were involved in the literature search, and AOA organized the methodology.

## Ethical Statement

Not Applicable.

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