



COMPARATIVE STUDY OF NUTRITIONAL COMPOSITION OF UNFORTIFIED AND FORTIFIED FERMENTED *PROSOPIS AFRICANA* SEEDS

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Abstract

Protein malnutrition is a significant challenge in many African countries. This contributes to inadequate nutritional security, resulting in alternative approaches to enhance plant-based foods' protein and mineral content and improve overall dietary health. *Prosopis africana* is one of the lesser-known perennial leguminous plants that play a key role in West African cuisine. Fermented *P. africana* seeds are said to be a rich source of protein, fatty acids, minerals, and some other vital nutrients. This study, therefore, aimed to examine the nutritional composition of fermented seeds of *P. africana* fortified with garlic and ginger. One (1 kg) of raw *P. africana* seeds and 500g of *Allium sativum* and *Zingiber officinale* were obtained, composited in different ratios (100:0, 90:10, 80:20, and 70:30), and fermented for 5 days. The fermented seeds were evaluated for microbial, proximate, and mineral composition, antioxidant properties, and vitamin C content. Among the formulations, 70% fermented seeds of *P. africana* fortified with 30% garlic showed the highest protein, fiber, and ash content, while 80% fermented seeds of *P. africana* fortified with 20% garlic exhibited the highest carbohydrate content and antioxidant properties. Zinc is the most abundant mineral found in all samples. 100% fermented seeds of *P. africana* have a major quantity; calcium, manganese, and copper were found in minute amounts. During the period of fermentation, the following microorganisms were isolated: *Micrococcus* sp, *Bacillus* sp, *Staphylococcus* sp, *Enterococcus* sp, *Proteus* sp, *Klebsiella* sp, *Lactobacillus* sp, and *Pseudomonas* sp in which *Bacillus* and *Mucor* species were predominant, alongside *Micrococcus*, *Lactobacillus*, and *Aspergillus niger*. These results demonstrate that fortifying fermented seeds of *P. africana* with ginger and garlic enhanced their nutritional profile, making it a healthier alternative to traditional seasoning salts and animal protein.

Keywords: *P. africana*, proximate, mineral composition, microorganisms, antioxidant properties

1. INTRODUCTION

Prosopis africana (Mesquite seeds) is one of the less well-known leguminous plants of about 45 species used as a condiment in the Middle Belt, North, and certain areas of Eastern and Southern Nigeria (Ojokoh and Eromosele, 2015; Balogun et al., 2017; Akpata et al., 2023). It is also found exclusively in tropical and subtropical regions of America, Southwest Asia, Senegal, and Ethiopia (Balogun and Oyeyiola, 2012; Balogun et al., 2017) holding a significant place in the culinary traditions of many communities in West Africa, serving as a flavor-enhancing agent (Popova and Mihaylova, 2019).

However, due to the high cost of animal protein, leguminous seeds became a growing interest as an alternative source of vegetable protein for both human and livestock consumption (Balogun et al., 2017). Although the seeds of *P. africana* are initially unsuitable for consumption, they become edible as food condiments through natural, spontaneous fermentation (Nwuche, 2013; Akpi et al., 2020). This makes them useful as seasoning in soups, stews, and other dishes, adding flavor and

variety to them i.e., a culinary enhancer (Fowoyo, 2017; Akpi et al., 2020). They are known to be protein-rich sources of essential nutrients. These condiments are integral to the human diet, significantly enhancing West African cuisine's sensory appeal and nutritional value (Egwin et al., 2013).

Fermented *P. africana* seeds are known by various native Nigeria names such as *Kiriya* (Hausa), *Kohi* (Fulani), *Sam chi lati* (Nupe), *Ayan* (Yoruba), *Kpaye* (Tiv), *Okpei* (Ibo), and *Okpehe/okpeye* (Idoma) (Fowoyo, 2017; Akpata et al., 2023). It is valued for its various health benefits and biologically active ingredients as reported by Agunwah et al. (2024). It offers a viable alternative to traditional animal-based ingredients, providing a tasty, high-protein substitute for fish or meat in soups and sauces, especially for low-income populations (Akpata et al., 2023), and also supports digestion and overall bodily functions due to its high nutrient content (Akpi et al., 2020; Ezekaibeya et al., 2020; Akpi et al., 2023). It is also known for its potent aroma and flavor-enhancing properties in various soups, rich in minerals, fatty acids, and essential nutrients such as phosphorus, potassium, and calcium, making

it a valuable addition to diets (Olaniran and Abiose, 2019; Achikanu *et al.*, 2020; Ezeocha *et al.*, 2022).

Furthermore, fermented *P. africana* seeds have been reported to help manage bacterial infections and support the development of the body's skeleton and cartilage (Ezeocha *et al.*, 2022). Most traditional diets generally comprise primarily staple foods, rich in calories but deficient in nutrients such as protein, vitamins, minerals, etc. with modest amounts of other meals based on availability and season (Balogun *et al.*, 2017) and this protein malnutrition is a significant challenge in many African countries, contributing to inadequate nutritional security (Fowoyo, 2017; Akpata *et al.*, 2023). Several research efforts have aimed to enhance the protein and mineral content of various plant-based foods including *P. africana* seeds through fermentation (Akpata *et al.*, 2023; Olaniran and Adediran, 2020).

Although, fermented *Prosopis africana* seeds have long been used as a condiment in soup, improving the overall nutritional composition of this condiment through fortification with garlic and ginger has not been explored. This present study seeks to evaluate the nutritional and microbiological composition of fermented *P. africana* seeds when fortified with garlic and ginger.

2. Materials and methods

2.1 Collection of Sample

Fresh samples of *P. africana* seeds (1kg), 500g of garlic (*Allium sativum*) and ginger (*Zingiber officinale*) respectively were obtained from were obtained from Owena market in Ondo town, placed in a clean plastic container, and transported to the laboratory for further analysis.

2.2 Preparation of Sample

Already weighed *P. africana* seeds, *A. sativum*, and *Z. officinale* were thoroughly cleaned and washed. The seeds of *P. africana* were boiled for 6 hours until they became soft and allowed to cool to room temperature (28 °C). The seeds were dehulled manually by pressing them between the palms of the hands to remove the cotyledon which was then re-boiled for 3 hours to further soften to hasten the fermentation process. The cotyledons were divided into seven (7) portions; first portion contained 200 g of *P. africana* seed only, the second, third, and fourth portions contained *P. Africana* seed fortified with *A. sativum* in different compositions (90:10, 80:20, 70:30). In the same vein, the remaining portions contained *P. africana* seeds fortified with *Z. officinale*, tightly wrapped with foil paper in a container and subjected to fermentation for 5 days using the traditional method (Akpata *et al.*, 2023; Agunwah *et al.*, 2024).

2.3 Determination of pH of fermented sample

The pH level of each sample was examined repeatedly at intervals throughout fermentation. One gram (1 g) of each

sample was weighed, mashed using a sterile mortar into a beaker, and dissolved in 9 ml of distilled water to form a slurry. The pH meter (Pye, Unicam pH meter, model 291) was standardized with buffer solution (pH 6.0) and the electrode was dipped into the slurry. The readings were recorded in duplicate (Eze *et al.*, 2014; Ibeabuchi *et al.*, 2014; Olaniran *et al.*, 2020).

2.4 Evaluation of microbial composition of fermented sample

2.4.1 Isolation and identification of isolated bacteria

Each sample (fortified and unfortified) was investigated for microorganisms responsible for its fermentation. A 1-gram sample of each was homogenized into 9 mL of sterile distilled water as stock, mixed thoroughly by agitation for 2 – 3 minutes, and a serial 10-fold dilution was done under aseptic conditions. A 0.1 ml of each sample (aliquot) of 10^{-6} dilution was inoculated into a prepared Nutrient Agar and MRS (de Mann, Rogosa and Sharpe), agar plate using the pour plate method. The plates were incubated aerobically and anaerobically at 30 °C for 15-24 hours in duplicates. This was done for the period of fermentation consecutively. The cultures were then purified successively to determine their morphological and biochemical characteristics according to the assay method of Obidi *et al.* (2023). The morphology of each isolate was observed; biochemical tests were also carried out. The pure cultures were stored on agar slants at 4 °C in a refrigerator

2.4.2 Isolation and identification of isolated fungi

Following the procedure stated in 2.4.1, an aliquot (0.1 ml) of (10^{-3}) dilution was inoculated into Potato dextrose agar (PDA) using the plate method; this was done for the period of fermentation consecutively. The asexual and sexual reproductive structures, like sporangia, conidia heads, ascospores, and vegetative mycelium of the fungal isolates, were observed microscopically using the culture slide method and the characteristics were compared with the characteristics of reference organisms (Kumar *et al.*, 2018).

2.5 Evaluation of the proximate composition of fermented samples

Each sample (fortified and unfortified) was evaluated for ash, crude fiber, fat, moisture, and protein content according to the standard method reported by Akpata *et al.* (2023); Yepto and Ajungla (2023).

2.5.1 Moisture content

The moisture content of each sample was determined by using the direct oven drying method. Clean and dried crucibles were labelled accordingly and weighed using a digital weighing balance, and their respective weights were recorded. Each sample (1 g) was weighed into the pre-weighed crucible and

transferred into the oven at 105 °C for 3 hours. After drying, the samples were allowed to cool before weighing. This process was repeated until a constant weight was achieved and recorded as the percentage moisture content.

2.5.2 Fat content

The oven-dried samples used to determine the moisture content were weighed, tightly packed, and placed in a Soxhlet extractor. Extraction was carried out with n-hexane (for defatting) at 40-60 °C under reflux. After extraction, the extracts were then placed inside an oven at 100°C for 1 hour and later allowed to cool in the desiccator, and the weight was recorded. This process continued for 5 days.

2.5.3 Protein content

During the period of fermentation, one gram (1 g) of each sample was ground with a porcelain mortar and added to 50 mL of distilled water, which was left for 1 hour. 2.5 µL was drawn from the mixture with a pipette. A 0.5 µL of reagent C (NaOH + NaCO₃), reagent B (Copper solution), and 0.8 mL of distilled water were added to the mixture and fixed for another 1 hour. The turbidity (absorbance) of the mixture was determined in a covet glass slide using a spectrometer (model UV1902).

2.5.4 Crude fiber content

Each sample (2.5 g) was measured using a weighing balance and pounded with a porcelain mortar then transferred into a 200 mL conical flask. 100 mL of H₂SO₄ (0.2 M) was dispensed into each sample and subjected to boiling for 1 hour which was then filtered using filter paper. 100 mL of KOH (1.125 M) was then added to the residue and also heated for 1 hour which was also filtered to obtain the crude fiber. The residue was then transferred into a crucible and oven dried and the weight was determined.

2.5.5 Ash content

One gram (1 g) of each sample was measured into clean, dried, pre-weighed crucibles. The organic matter was burned off using flame until the sample became charred. The crucible was then transferred to a muffle furnace set at 550°C. This process continued until a light grey color of white ash was obtained. The crucibles were then allowed to cool, weighed, and recorded.

2.6 Determination of the mineral composition of the fermented sample

The mineral constituents of each sample (fortified and unfortified) were determined following the Aremu et al. (2015) procedure, modified by Olaniran and Abiose (2019). The ash content obtained from the samples was liquefied in 10 mL of 2 M HNO₃, boiled for 5 min, filtered into a volumetric flask, and made up with distilled water to 50mL. The concentrations of manganese, iron, zinc, potassium, calcium, and copper were

determined in the sample using an atomic absorption spectrophotometer (Model UV1902).

2.7 Determination of vitamin C content of fermented sample

The vitamin C content of each sample (fortified and unfortified) was evaluated according to standard methods stated by Carazo et al. (2021); Akpata et al. (2023). One gram (1 g) of each sample was added to 1g of fat and 240 units of vitamin C, thoroughly mixed with 30 ml of absolute alcohol and 3 ml of 5% potassium hydroxide. The mixture was then gently boiled under reflux for 30 minutes in a stream of oxygen-free nitrogen. Afterward, the mixture was rapidly cooled, and 30 ml of water was added. The solution was then transferred to a separator, washed with three 50 ml portions of ether, and the vitamins were extracted by shaking for 1 minute. Once the layers had completely separated, the lower layer was discarded, and the extract was washed with four 50 ml portions of water. Special care was taken during the first two washes to avoid emulsion formation. The washed extract was evaporated to approximately 5 ml, and the remaining ether was removed in a stream of nitrogen at room temperature. Finally, the residue was dissolved in sufficient isopropyl alcohol to create a solution containing 9-15 units per ml, and the extinctions were measured at 300, 310, 325, and 334 nm, along with the wavelength of maximum absorption.

2.8 Determination of antioxidant properties of fermented samples

The antioxidant properties of each sample (fortified and unfortified) were determined using the DPPH assay method of Ozioko et al. (2020) modified by Yephtho and Ajungla, (2023). One milliliter (1 ml) of each sample extract was mixed with 1 ml of the 0.4 mM methanolic solution of the 2,2-diphenyl-1-picrylhydrazyl the mixture was left in the dark for 30 minutes, and measured with a spectrophotometer (absorbance at 516 nm).

2.9 Statistical analysis

Experimental results were expressed as means ± standard deviation and data was subjected to one-way analysis of variance (ANOVA) using SPSS version 25.0.

3. Results and discussion

The pH of the fermented seeds of *P. africana* (fortified and unfortified) was alkaline, during the first three days of fermentation, there was a stable pH between 6.30 and 6.45 (Fig. 3.1). However, at day 3, there was a progressive rise in pH level across the samples. The rise in pH level could also be due to the high proteinase activity of microorganisms (Fowoyo, 2017), high protein content in the fermented seed resulting from the elimination of the species responsible for acid fermentation during processing which encourages a non-acid fermentation

(Ibeabuchi et al., 2014). It could also be attributed to the production of ammonia or other alkaline compounds due to the breakdown of proteins and other organic materials by microorganisms resulting from the elimination of the species responsible for acid fermentation during processing which encourages a non-acid fermentation (Balogun et al., 2017).

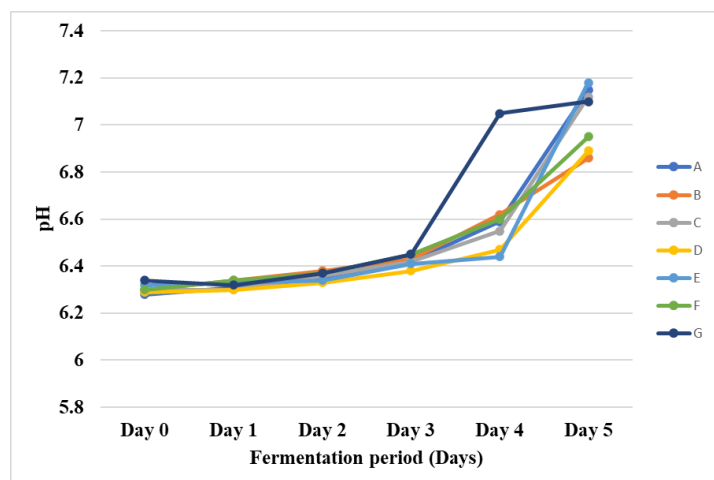


Fig 3.1: pH level of fermented *P. africana* seed during fermentation

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Diverse group of microorganisms were isolated from the fermented seeds of *P. africana* during the fermentation period. The total bacteria count (TBC) of the fermented condiment in **Table 3.1** revealed that sample A and B had the highest bacterial count at day 1 (24.6 ± 1.07 and 21.2 ± 0.17) respectively, at the end of fermentation, sample C, D, and E has the lowest count (2.8 ± 0.02). At day 1, sample G had the lowest count (3.7 ± 0.05).

Table 3.1: Total bacterial count (TBC) of fermented *P. africana* seeds

Sample	Fermentation period/Bacterial count ($\times 10^6$ (CFU/g))				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	5.7 ± 0.03^c	19.5 ± 0.23^b	24.6 ± 1.07^a	9.9 ± 0.12^d	10.6 ± 0.77^c
B	5.7 ± 0.03^d	14.2 ± 0.19^b	21.2 ± 0.17^a	4.8 ± 0.22^e	6.4 ± 0.07^c
C	4.2 ± 0.03^c	9.5 ± 0.11^b	13.2 ± 0.02^a	4.8 ± 0.22^c	2.8 ± 0.02^d
D	4.2 ± 0.41^c	9.5 ± 0.07^b	13.2 ± 0.07^a	4.8 ± 0.22^c	2.8 ± 0.01^d
E	4.5 ± 0.03^c	9.3 ± 0.04^b	14.6 ± 0.05^a	4.8 ± 0.07^c	2.8 ± 0.01^d
F	4.8 ± 0.07^c	14.0 ± 0.05^b	19.2 ± 1.03^a	8.9 ± 0.03^d	9.7 ± 0.07^c
G	3.7 ± 0.05^c	11.6 ± 0.03^b	15.0 ± 0.05^a	9.9 ± 0.10^c	7.0 ± 0.03^d

Key: A-100% okpei only, B-90% okpei+ 10% ginger, C-80% okpei + 20% ginger, D- 70% okpei + 30% ginger, E-90% okpei+ 10% garlic, F-80% okpei + 20% garlic, G-70% okpei + 30% garlic, values of mean are of duplicate determination \pm SD, mean in the same column with different superscripts are significantly different at ($p < 0.05$).

Samples E, F, and G had the highest total fungi count at day 1 while sample A had the lowest count. Sample C had the highest colony count at day 3 (10 ± 0.18) as shown in Table 3.2. After fermentation, no bacterial growth was observed in sample A, B, F, and G at day 4 and 5.

Table 3.2: Total fungal count (TFC) of fermented *P. africana* seeds

Sample	Fermentation period/Fungal count (CFU/g $\times 10^3$)				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	0.2 ± 0.00^c	3.0 ± 0.00^a	3.2 ± 0.40^a	NG	NG
B	1.2 ± 0.01^c	3.0 ± 0.00^b	6.9 ± 0.20^a	NG	NG
C	1.2 ± 0.01^c	5.3 ± 0.10^b	10 ± 0.18^a	3.5 ± 0.20^c	2.1 ± 1.00^d
D	1.0 ± 0.00^d	2.3 ± 0.12^c	6.8 ± 0.20^a	3.5 ± 0.20^b	2.1 ± 1.00^c
E	2.0 ± 0.03^d	2.6 ± 0.04^d	4.9 ± 0.51^b	5.9 ± 1.30^a	3.3 ± 1.70^c
F	2.0 ± 0.03^a	0.3 ± 0.03^c	1.8 ± 0.04^{ab}	NG	NG
G	2.0 ± 0.30^{ab}	0.3 ± 0.03^c	1.8 ± 0.04^b	NG	NG

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic, NG – No growth, values of mean are of duplicate determination \pm SD, mean in the same column with different superscripts are significantly different at ($p < 0.05$).

Fig. 3.2 and Fig. 3.3 revealed the percentage occurrences of the isolated microorganisms in each sample during the fermentation period. *Micrococcus* sp was isolated in sample (A, F, and G), only 1% was isolated at day 1, as the fermentation progresses 19.6% was isolated at day 2, day 3 (22.7%), day 4 (31.0%) and day 5 (25%). *Bacillus* sp and *Lactobacillus* sp were isolated in all samples; 6.8% of *Bacillus* sp was isolated at day 1 increasing to 12.8% after fermentation while 35.8% of *Lactobacillus* sp was isolated at day 1 and at day 4, it was terminated. 44.1% *staphylococcus* sp was isolated in sample (A, C, and D) from day 1 to day 3 after which was terminated, 22% *Klebsiella* sp were isolated in sample (A, B, and F), 14.3% *Enterococcus* sp were isolated from sample (A and G), 11.8% *Proteus* sp were isolated from sample (B and E) and 25% *Pseudomonas* sp were isolated from sample G only after the fermentation period. In Fig. 3.3, the total fungi isolated from fermented condiment include: 6.2% *Aspergillus niger* isolated in sample (A, B, and C) at day 1, 23.7% at day 2 and was terminated thereafter, 21.2% and 65.9% *Saccharomyces* sp (B, C, and D) at day 1 and 4. *Mucor* sp were isolated in all samples from day 2 to day 5. 4.1% *Rhizopus* sp were isolated in sample (E, F, and G) at day 1, at day 2 the total percentage occurrence in all sample increases to 12.7% and was terminated at day 3.

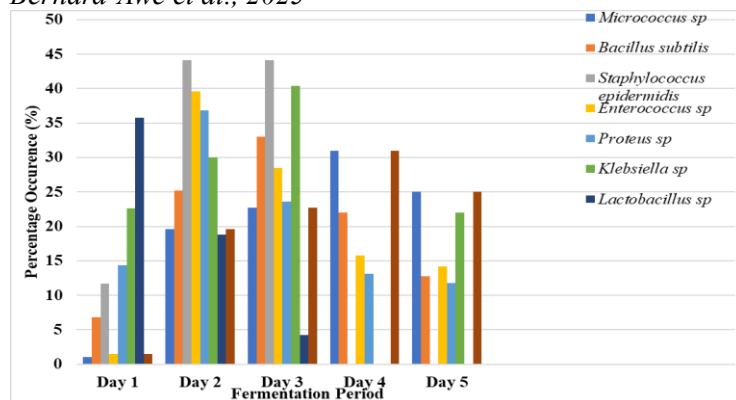


Fig. 3.2: Percentage occurrence of isolated bacteria from fermented *P. africana* seeds

During fermentation, the occurrence of microorganisms at different stage of fermentation showed that the predominant organism involved in the fermentation were *Bacillus* sp, *Micrococcus* sp, and *Lactobacillus* sp. *Staphylococcus* sp, *Klebsiella* sp, and *Pseudomonas* sp was present up to day 3. This result agrees with Balogun and Oyeyiola, (2011) investigation. *Bacillus* sp were more predominant in all the samples, this is because they are associated with fermented legume seeds as reported by Fowoyo, (2017), the presence of these organisms may lead to breakdown of protein leading to an increased proteinase activity contributing to the development of texture and flavour (Ukaoma et al., 2019). *A. niger* was isolated from sample A, B, C, *Mucor* in C, D, E, *Saccharomyces* sp in B, C, D and *Rhizopus* sp in E, F, G respectively. It was observed that all fungal growth were terminated at day 3 except *Mucor* sp that was dominant throughout the fermentation period. According to Fowoyo (2017), fermented *P. africana* seeds (okpei) is rich in phytochemical compounds such as saponin which particularly help to reduce protein digestibility. It was also reported to possess antifungal properties; which help to prevent proliferation of fungi.

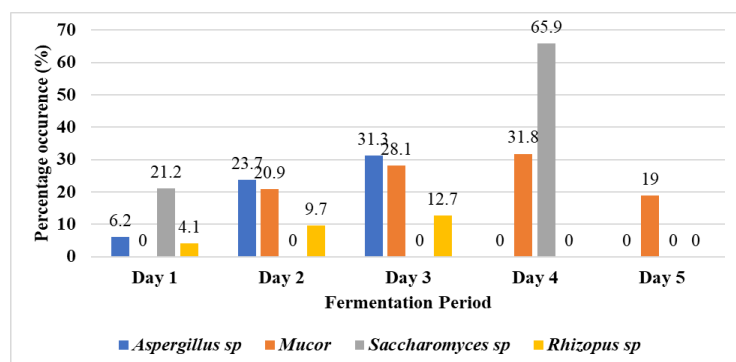


Fig. 3.3: Percentage occurrence of isolated fungi from fermented *P. africana* seeds

During the period of fermentation, the proximate evaluation of the fermented *P. africana* seeds showed that on day 1, 90% fermented seeds of *P. africana* with 10% ginger exhibited the highest moisture content ($67.28 \pm 0.12\%$), while sample A had the lowest ($57.14 \pm 0.01\%$) as revealed in Table 3.4. 70% fermented seeds of *P. africana* with 30% ginger recorded the highest moisture content on day 2 ($67.45 \pm 0.01\%$), followed closely by 80% fermented seeds of *P. africana* with 20% ginger ($67.73 \pm 1.19\%$) and 90% fermented seeds of *P. africana* with 10% garlic ($67.45 \pm 0.20\%$). In contrast, 70% fermented seeds of *P. africana* with 30% garlic exhibited the lowest moisture content during the fermentation period, declining to $49.91 \pm 1.16\%$ on day 4. After fermentation, 80% fermented seeds of *P. africana* with 20% ginger had the highest moisture content ($62.09 \pm 0.42\%$), closely followed by 70% fermented seeds of *P. africana* with 30% ginger ($59.46 \pm 1.10\%$). However, 100% fermented seeds of *P. africana* had the lowest moisture content at the end of the fermentation period ($54.73 \pm 1.56\%$). In Balogun, (2012) and Fowoyo, (2017) findings, the moisture content of the fermented condiment ranged from 5.13 – 8.64. In this study, the moisture content of all samples after fermentation ranged from 54.73 – 62.09 and this agrees with Olaniran and Abiose, (2019) and Ogbu (2024). At the end of fermentation, 80% fermented seeds of *P. africana* with 20% ginger had a higher moisture content (62.09%) than other samples. This is in agreement Olaniran and Abiose, (2019) findings.

Table 3.4: Moisture content of fermented *Prosopis africana* seeds during fermentation

Sample	Moisture composition % (Mean ± SD)				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	57.14 ± 0.01^c	59.73 ± 0.23^c	58.00 ± 0.23^d	56.81 ± 1.07^d	54.73 ± 1.56^f
B	67.28 ± 0.12^a	62.44 ± 1.57^b	60.68 ± 1.55^c	66.45 ± 1.12^a	55.06 ± 5.46^e
C	62.81 ± 0.21^b	67.73 ± 1.19^a	65.91 ± 1.17^a	56.72 ± 0.06^d	62.09 ± 0.42^a
D	59.43 ± 0.04^d	67.45 ± 0.01^a	65.63 ± 0.01^a	59.78 ± 0.14^b	59.46 ± 1.10^b
E	62.33 ± 0.11^b	67.45 ± 0.20^a	63.80 ± 0.19^b	56.10 ± 0.68^d	55.98 ± 4.07^e
F	57.64 ± 0.13^c	59.80 ± 0.28^c	58.06 ± 0.28^d	57.71 ± 0.05^c	57.30 ± 2.77^c
G	57.66 ± 0.10^c	62.18 ± 0.5^b	60.42 ± 0.57^c	49.91 ± 1.16^e	56.15 ± 0.58^d

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Note: mean in the same column with different superscripts are significantly different at ($p < 0.05$).

Table 3.3: Microscopic, biochemical and sugar fermentation profile of bacteria isolated from fermented *P. africana* seeds

Morphological Characteristics	Biochemical tests							Sugar Fermentation					
	Gram Reaction	Catalase	Citrate Utilization	MR	VP	Oxi	Coagulase	Indole	Glu	Fru	Suc	Mal	Probable Organism
Small, circular whitish colonies	+	-	+	-	-	-	-	-	A	A	A	A	Lactobacillus sp
Smooth, opaque, pale-yellowish colonies	+	+	-	+	+	-	+	+	AG	A	A	A	Staphylococcus epidermidis
Translucent with visible green pigments colonies	-	+	-	+	+	+	-	+	AG	AG	A	AG	Pseudomonas sp
Smooth, pink creamy mucoid colonies	-	+	+	-	+	-	-	-	+	+	+	+	Klebsiella sp
Large swamy creamy moist and flat colonies	-	+	+	+	-	-	-	-	+	+	+	+	Proteus sp
Entire, raised whitish, colonies	+	-	+	+	+	-	+	-	+	+	+	AG	Enterococcus sp
Circular, smooth, creamy yellowish colonies	+	+	-	+	+	+	-	-	-	-	-	-	Micrococcus sp
Dry creamy mucoidal, circular, slightly raised colonies	+	+	+	-	+	+	-	-	A	A	A	A	Bacillus subtilis

Key: A- Acid production, AG- Acid and gas production, + (positive), - (negative), MR- Methyl red, VP- Vogues Proskauer, Oxi-Oxidase, Glu-Glucose, Fru-Fructose, Suc-Sucrose, Mal-Maltose

The ash composition of all samples during fermentation revealed that sample A had the lowest ash content by day 5 (1.97%), while 90% fermented seeds of *P. africana* with 10% garlic recorded the highest value at 4.18% on day 3 (**Table 3.5**). 70% fermented seeds of *P. africana* with 30% garlic consistently exhibited low ash content throughout the fermentation period, with 1.92% on day 5. In contrast, 80% fermented seeds of *P. africana* with 20% garlic had a high initial ash content of 3.21% on day 1, which declined over time. At the end of fermentation, 30% fermented seeds of *P. africana* fermented with 20% ginger had higher ash content than the other proportions (2.19%), followed by 80% fermented seeds of *P. africana* fermented with 20% garlic (2.18%) after fermentation. The presence of ash indicated that the condiment is rich in minerals (Fowoyo, 2017). There was a progressive increase in fat content of 70% fermented seeds of *P. africana* fermented with 30% ginger rising from (3.33 – 6.05%) throughout fermentation as revealed in Table 3.6 This aligned with Ozioko et al. (2020) report, which could be attributed to the presence of oil (Olaniran and Adeniran, 2020).

Table 3.5: Ash content of fermented *Prosopis africana* seeds during fermentation

Sample	Ash composition % (Mean ± SD)				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	2.51±0.06 ^b	2.37±0.06 ^b	2.37±0.06 ^c	2.22±0.06 ^b	1.97±0.03 ^b
B	2.41±0.14 ^b	2.26±0.01 ^b	2.26±0.01 ^c	2.12±0.01 ^b	2.16±0.05 ^a
C	2.11±0.16 ^b	1.96±0.16 ^c	1.96±0.16 ^d	1.82±0.16 ^c	1.87±0.10 ^b
D	2.33±0.10 ^b	2.18±0.10 ^b	2.18±0.10 ^c	2.04±0.10 ^b	2.19±0.17 ^a
E	3.33±0.01 ^a	2.18±0.14 ^b	4.18±0.14 ^a	3.03±0.14 ^a	2.07±0.24 ^a
F	3.21±0.02 ^a	3.06±0.02 ^a	3.06±0.02 ^b	2.91±0.01 ^b	2.18±0.02 ^a
G	2.72±0.01 ^b	2.58±0.01 ^b	2.58±0.04 ^c	2.43±0.00 ^b	1.92±0.07 ^b

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Table 3.6: Fat content of fermented *Prosopis africana* seeds during fermentation

Sample	Fat composition % (Mean ± SD)				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	2.36±0.09 ^b	2.38±0.09 ^c	2.52±0.09 ^b	2.83±0.11 ^b	4.36±0.17 ^c
B	2.93±0.68 ^b	2.95±0.22 ^b	3.10±0.22 ^a	3.48±0.24 ^a	5.35±0.38 ^b
C	2.02±0.59 ^b	2.03±0.59 ^c	2.18±0.57 ^b	2.45±0.67 ^b	3.76±1.03 ^d
D	3.33±0.47 ^a	3.36±0.47 ^a	3.50±0.47 ^a	3.94±0.53 ^a	6.05±0.82 ^a
E	2.04±0.68 ^b	2.06±0.69 ^c	2.20±0.69 ^b	2.47±0.77 ^b	3.80±1.19 ^d
F	2.23±0.01 ^b	2.24±0.01 ^c	2.39±0.01 ^b	2.68±0.01 ^b	4.13±0.02 ^c
G	2.77±0.17 ^b	2.80±0.17 ^b	2.94±0.17 ^b	3.30±0.10 ^a	5.08±0.31 ^b

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Note: mean in the same column with different superscripts are significantly different at ($p < 0.05$).

There was a progressive increase in fiber content across all samples during the fermentation period as revealed in **Table 3.7**. 80% fermented seeds of *P. africana* with 20% ginger had the lowest fiber content on day 1 (2.43±0.14), which steadily increased to 5.77±0.34 by day 5. 90% fermented seeds of *P. africana* with 10% garlic exhibited the most significant increase in fiber content, peaking at 9.13±0.12 by the end of fermentation, followed by 80% fermented seeds of *P. africana* with 20% garlic (8.54±0.04). The other samples displayed moderate but consistent increases in fiber content. The protein content of the samples progressively increased throughout the fermentation period as shown in **Table 3.8**. 100% fermented seeds of *P. africana* had the highest protein content, increasing significantly from 4.67±0.16 on day 1 to 23.94±3.27 by the end of fermentation. Similarly, 90% fermented seeds of *P. africana* with 10% garlic demonstrated a high initial protein content (8.77±0.27) that increased to 22.73±0.24, although it was slightly lower than that of 100% fermented seeds of *P. africana*. 80% fermented seeds of *P. africana* with 20% ginger and 70% fermented seeds of *P. africana* with 30% ginger exhibited similar protein levels at the end of fermentation, with values of 19.88±1.0 and 19.68±0.78, respectively.

Table 3.7: Fiber content of fermented *Prosopis africana* seeds during fermentation

Sample	Fiber composition % (Mean \pm SD)				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	2.80 \pm 0.19 ^c	3.59 \pm 0.05 ^b	4.56 \pm 0.06 ^c	6.36 \pm 0.08 ^c	6.95 \pm 0.08 ^c
B	2.87 \pm 0.14 ^c	2.89 \pm 0.01 ^c	3.72 \pm 0.01 ^d	5.25 \pm 0.01 ^d	5.84 \pm 0.01 ^d
C	2.43 \pm 0.01 ^c	2.85 \pm 0.21 ^c	3.67 \pm 0.25 ^d	5.18 \pm 0.34 ^d	5.77 \pm 0.34 ^d
D	3.31 \pm 0.01 ^b	3.47 \pm 0.09 ^b	4.42 \pm 0.12 ^c	6.17 \pm 0.15 ^c	6.76 \pm 0.15 ^c
E	4.77 \pm 0.14 ^a	4.94 \pm 0.07 ^a	6.21 \pm 0.09 ^a	8.54 \pm 0.12 ^a	9.13 \pm 0.12 ^a
F	3.91 \pm 0.02 ^b	4.57 \pm 0.02 ^a	5.76 \pm 0.03 ^b	7.95 \pm 0.02 ^b	8.54 \pm 0.04 ^b
G	3.25 \pm 0.13 ^b	3.20 \pm 0.35 ^b	4.63 \pm 0.42 ^c	5.73 \pm 0.28 ^d	6.32 \pm 0.56 ^c

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Note: mean in the same column with different superscripts are significantly different at ($p < 0.05$).

Table 3.8: Protein content of fermented *Prosopis africana* seeds during fermentation

Sample	Protein composition % (Mean \pm SD)				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	4.67 \pm 0.16 ^d	7.95 \pm 0.61 ^f	11.59 \pm 0.25 ^b	17.85 \pm 0.32 ^c	23.94 \pm 3.27 ^a
B	4.60 \pm 0.27 ^d	8.36 \pm 0.12 ^c	8.26 \pm 0.03 ^d	14.58 \pm 0.58 ^c	18.36 \pm 0.18 ^d
C	6.01 \pm 0.12 ^c	9.04 \pm 1.02 ^d	12.48 \pm 1.36 ^a	20.71 \pm 0.38 ^a	19.88 \pm 1.06 ^c
D	7.66 \pm 1.18 ^b	12.95 \pm 0.84 ^a	11.01 \pm 0.47 ^b	17.67 \pm 0.76 ^c	19.68 \pm 0.78 ^c
E	8.77 \pm 0.27 ^a	11.04 \pm 1.18 ^b	11.26 \pm 0.24 ^b	18.63 \pm 0.37 ^b	22.73 \pm 0.24 ^b
F	2.06 \pm 0.66 ^c	8.39 \pm 1.57 ^c	11.90 \pm 0.84 ^b	16.53 \pm 0.03 ^d	15.84 \pm 0.12 ^c
G	8.63 \pm 0.23 ^a	10.70 \pm 1.69 ^c	10.76 \pm 1.42 ^c	14.58 \pm 0.10 ^c	13.11 \pm 0.07 ^f

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Note: mean in the same column with different superscripts are significantly different at ($p < 0.05$).

Table 3.9 shows that there was a significant decrease in carbohydrate levels across all samples as fermentation progressed. 100% fermented seeds of *P. africana* and 80% fermented seeds of *P. africana* with 20% garlic initially had relatively high carbohydrate content (30.53 \pm 0.21 and 30.96 \pm 0.7, respectively), which declined to 8.06 \pm 5.05 and 12.01 \pm 2.61 after fermentation. However, 70% fermented seeds of *P. africana* with 30% garlic maintained a higher carbohydrate level by day 5 (17.41 \pm 0.99) compared to 100% fermented seeds of *P. africana* (13.23 \pm 5.05) and 80%

fermented seeds of *P. africana* with 20% garlic (12.01 \pm 2.61). 70% fermented seeds of *P. africana* with 30% ginger exhibited the lowest carbohydrate content among all samples after fermentation. Generally, the fortified condiment exhibited a significant higher fiber content than the unfortified condiment at the end of fermentation as reported by (Balogun and Oyeyiola, 2012). However, this result differs from Okafor *et al.* (2018); Olaniran and Abiose, (2019) as lower level of fiber content was reported. 100% fermented seeds of *P. africana* had the highest protein content (23.94%) after fermentation followed by 90% fermented seeds of *P. africana* with 10% garlic (22.73%). The unfortified condiment had a lower carbohydrate content after fermentation period (8.06%), its presence indicates the digestibility ability of the product while 70% fermented seeds of *P. africana* with 30% garlic had the highest amount (17.41%).

Table 3.9: Carbohydrate content of fermented *Prosopis africana* seeds during fermentation

Sample	Carbohydrate composition % (Mean \pm SD)				
	Day 1	Day 2	Day 3	Day 4	Day 5
A	30.53 \pm 0.21 ^a	23.99 \pm 0.59 ^a	20.96 \pm 0.25 ^b	13.92 \pm 1.00 ^b	8.06 \pm 5.05 ^d
B	19.91 \pm 0.01 ^d	21.09 \pm 1.49 ^b	21.98 \pm 1.38 ^a	8.12 \pm 1.93 ^f	13.23 \pm 5.22 ^b
C	24.63 \pm 0.67 ^b	16.39 \pm 0.81 ^d	13.80 \pm 1.52 ^c	13.12 \pm 1.62 ^b	6.63 \pm 0.84 ^c
D	23.45 \pm 1.69 ^c	10.59 \pm 1.32 ^f	13.25 \pm 0.26 ^c	10.40 \pm 0.14 ^c	5.86 \pm 0.16 ^f
E	18.11 \pm 0.01 ^c	13.18 \pm 1.45 ^c	12.34 \pm 0.50 ^f	11.23 \pm 0.82 ^d	6.29 \pm 5.64 ^c
F	30.96 \pm 0.7 ^a	21.93 \pm 1.27 ^b	18.83 \pm 1.13 ^c	12.21 \pm 0.02 ^c	12.01 \pm 2.61 ^c
G	24.97 \pm 0.39 ^b	18.55 \pm 1.29 ^c	17.47 \pm 1.10 ^d	24.03 \pm 1.24 ^a	17.41 \pm 0.99 ^a

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Note: mean in the same column with different superscripts are significantly different at ($p < 0.05$).

Potassium, calcium, copper, iron, manganese, zinc, and lead was detected in varying and appreciable quantity in the fermented samples during the fermentation period as shown in table 4.0. Zinc was the most abundant mineral found in all samples. 100% fermented seeds of *P. africana* had a major quantity while calcium, manganese, and copper were found in minute quantities. 70% fermented seeds of *P. africana* with 30% garlic have the most abundant level of calcium, manganese, and copper. 80% fermented seeds of *P. africana* with 20% ginger is abundant in potassium which is in agreement with Fowoyo (2017) findings. At the end of fermentation, 70% fermented seeds of *P. africana* with 30% garlic was rich in potassium, zinc, and calcium while the unfortified fermented seeds of *P. africana* was rich in iron.

Calcium is a good element needed for bone development thus, its presence in the condiment is good. Zinc was also found in an appreciable quantity in 70% fermented seeds of *P. africana* with 30% garlic than (Fowoyo, 2017) which ranged 0.142 – 0.172 mg/g. This is desirable as it is known to aid digestion and body functions. Copper was also detected in all the samples which is a trace element that serves as a co-factor and is required for enzyme function. However, the quantity of copper found in unfortified fermented seeds of *P. africana* produced was reduced at the end of fermentation (1.2 – 0.9 mg/g) whereas the fortified fermented seeds of *P. africana* with ginger and garlic in various composition had a constant value of 0.6 mg/g and 0.8 mg/g respectively. Lead was present in a minimal amount corresponding with Olajide et al. (2021). However, this differs from the report of Balogun (2012) and Fowoyo (2017), which recorded amount of Lead present as BDL (below detection limit). Zinc was the most abundant mineral with relatively high quantity after fermentation. However, Fowoyo, (2017) and Popova and Mihaylova, (2019) reported potassium to be the most abundant mineral present in fermented seeds of *P. africana*.

The vitamin C composition of each sample during the period of fermentation as shown in Fig. 3.4 revealed a progressive increase throughout the fermentation process at significant levels, with samples A, D, E, and G showing the most significant changes by the end of the period. However, 70% okpei with 30% garlic exhibited higher levels than the other samples (94.02%) while the amount in 100% okpei was relatively low throughout fermentation. Contrary to Balogun and Oyeyiola, (2012) and Fowoyo, (2017), the values were below the detection limit.

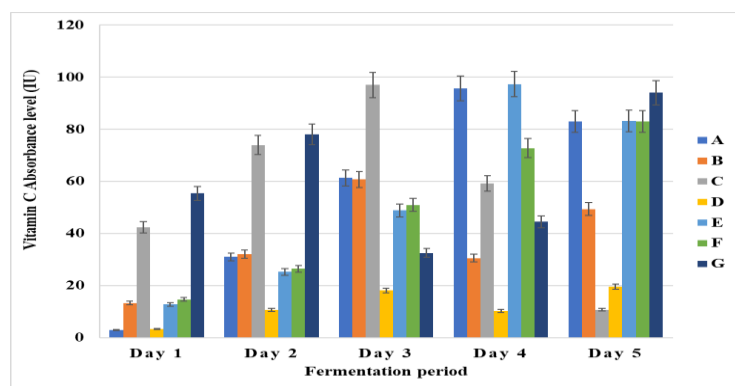


Fig. 3.4: Vitamin C content of fortified and unfortified fermented seeds of *P. africana* during fermentation

Key: A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

The antioxidant properties of fortified and unfortified fermented seeds of *P. africana* showed that the fortified fermented seeds of *P. africana* with ginger possessed high antioxidant properties during the period of fermentation as shown in Fig. 3.5 and 3.6, 80% fermented seeds of *P. africana* with 20% ginger had 27.9% at the beginning of fermentation and at the end 81.14% while 70% fermented seeds of *P. africana* with 30% ginger had 27.49% increasing to 91.25%. According to Ezeocha et al. (2022), fermented seeds of *P. africana* are said to be rich in antioxidant with garlic and ginger possessing good free radical scavenging abilities that can be used as radical inhibitors in considerable amount (Olaniran and Abiose, 2019). In this study, the fortified condiment with ginger showed stronger radical scavenging activity than the unfortified condiment and those fortified with ginger where as Asimi et al. (2013) reported garlic to possess stronger radical scavenging activity corresponding with Ryu et al. (2017). The percentage inhibition of DPPH free radical scavenging activity showed that 70% fermented seeds of *P. africana* with 30% ginger had the highest activity (91.25%) at the end of fermentation, while the percentage inhibition of ferric reducing acid property (FRAP) showed that 90% fermented seeds of *P. africana* with 10% garlic had the highest activity (2.59%) after fermentation aligning with (Asimi et al., 2013).

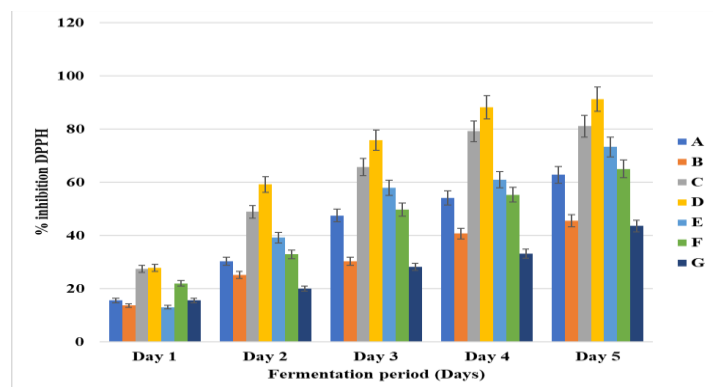


Fig 3.5: Percentage inhibition of DPPH free radical scavenging activity of fermented *P. africana* seeds during fermentation

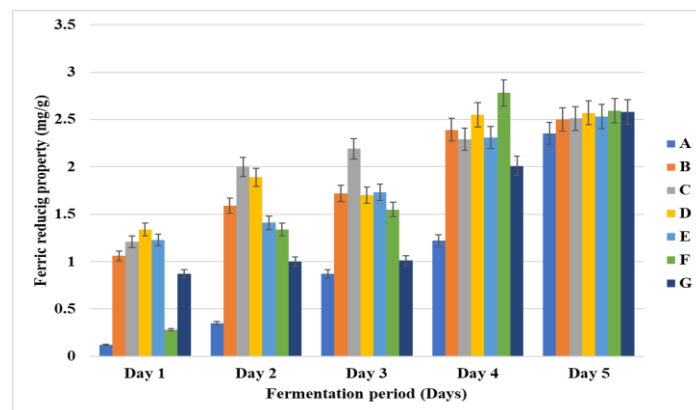


Fig 3.6: Percentage inhibition of ferric reducing acid property (FRAP) of fermented *P. africana* seeds during fermentation.

Table 3.9: Mineral composition of fermented *Prosopis africana* seeds during fermentation

Mineral Content	Sample	Fermentation Period (mg/g)				
		Day 1	Day 2	Day 3	Day 4	Day 5
K	A	0.19 ±0.01 ^a	0.20±0.01 ^a	0.36±0.01 ^b	0.40±0.01 ^c	0.35±0.02 ^{bs}
	B	0.25 ±0.00 ^a	0.30±0.01 ^b	0.40±0.02 ^{cd}	0.38±0.01 ^c	0.47±0.01 ^e
	C	0.29 ±0.00 ^a	0.35±0.01 ^b	0.42±0.01 ^{cd}	0.41±0.00 ^c	0.44±0.02 ^d
	D	0.26 ±0.01 ^a	0.36±0.01 ^b	0.42±0.01 ^c	0.40±0.01 ^c	0.36±0.00 ^b
	E	0.24 ±0.01 ^a	0.45±0.02 ^c	0.47±0.01 ^d	0.42±0.02 ^b	0.46±0.15 ^{cd}
	F	0.34 ±0.01 ^a	0.40±0.01 ^b	0.58±0.07 ^e	0.45±0.03 ^c	0.48±0.02 ^{cd}
	G	0.36 ±0.00 ^{ab}	0.33±0.00 ^a	0.43±0.01 ^d	0.39±0.01 ^{bc}	0.57±0.01 ^e
Ca	A	0.23 ±0.01 ^a	0.26±0.00 ^b	0.26±0.03 ^b	0.26±0.00 ^b	0.26±0.03 ^b
	B	0.39 ±0.04 ^b	0.35±0.02 ^a	0.47±0.00 ^c	0.40±0.01 ^b	0.42±0.04 ^b
	C	0.31 ±0.04 ^a	0.45±0.04 ^{cd}	0.34±0.03 ^{ab}	0.41±0.02 ^c	0.35±0.01 ^b
	D	0.32 ±0.03 ^{ab}	0.30±0.03 ^a	0.31±0.00 ^a	0.35±0.04 ^c	0.34±0.07 ^{bc}
	E	0.29 ±0.02 ^b	0.34±0.01 ^c	0.28±0.01 ^b	0.20±0.00 ^a	0.39±0.14 ^d
	F	0.34 ±0.01 ^a	0.45±0.03 ^b	0.52±0.02 ^{cd}	0.53±0.02 ^d	0.50±0.02 ^c
	G	0.46 ±0.01 ^b	0.24±0.00 ^a	0.49±0.04 ^{bd}	0.45±0.01 ^b	0.52±0.04 ^c
Cu	A	0.47 ±0.01 ^c	0.32±0.02 ^{bd}	0.29±0.02 ^b	0.30±0.01 ^b	0.24±0.07 ^a
	B	0.46 ±0.01 ^b	0.43±0.01 ^a	0.84±0.01 ^c	1.04±0.19 ^f	0.50±0.01 ^d
	C	0.29 ±0.01 ^a	0.53±0.01 ^d	0.48±0.02 ^c	1.32±0.12 ^e	0.31±0.07 ^b
	D	0.46 ±0.41 ^d	0.40±0.01 ^b	0.41±0.04 ^c	0.30±0.00 ^a	0.65±0.02 ^c
	E	0.37 ±0.01 ^a	0.48±0.01 ^b	0.82±0.01 ^d	0.92±0.01 ^c	0.57±0.23 ^c
	F	0.42 ±0.01 ^b	0.44±0.02 ^b	0.54±0.02 ^c	0.68±0.00 ^d	0.28±0.07 ^a
	G	0.19 ±0.00 ^a	0.41±0.02 ^d	0.39±0.01 ^c	0.33±0.01 ^b	0.83±0.01 ^e
Fe	A	1.24 ±0.00 ^d	1.23±0.23 ^d	0.98±0.02 ^c	0.58±0.00 ^a	0.91±0.02 ^b
	B	0.65 ±0.00 ^a	1.00±0.02 ^d	1.61±0.01 ^e	1.86±0.43 ^f	0.69±0.02 ^{bc}
	C	0.91 ±0.00 ^b	1.09±0.37 ^c	1.13±0.00 ^d	1.22±0.22 ^e	0.72±0.02 ^a
	D	1.03 ±0.01 ^c	0.86±0.10 ^b	1.02±0.00 ^c	1.00±0.00 ^c	0.49±0.00 ^a
	E	0.84 ±0.01 ^a	1.01±0.05 ^b	1.47±0.01 ^d	1.24±0.12 ^c	0.84±0.37 ^a

	D	1.03 ±0.01^c	0.86±0.10^b	1.02±0.00^c	1.00±0.00^c	0.49±0.00^a
Mn	E	0.84 ±0.01 ^a	1.01±0.05 ^b	1.47±0.01 ^d	1.24±0.12 ^c	0.84±0.37 ^a
	F	0.84 ±0.01 ^b	1.00±0.05 ^c	1.02±0.01 ^{cd}	0.99±0.01 ^c	0.69±0.01 ^a
	G	1.03 ±0.14 ^c	1.22±0.33 ^c	0.85±0.01 ^b	0.73±0.01 ^a	0.86±0.02 ^b
	A	0.29 ±0.01 ^{bc}	0.30±0.00 ^c	0.31±0.00 ^c	0.31±0.02 ^c	0.20±0.01 ^a
	B	0.26 ±0.00 ^b	0.21±0.02 ^a	0.40±0.07 ^d	0.40±0.01 ^d	0.38±0.01 ^c
	C	0.19 ±0.01 ^a	0.38±0.01 ^d	0.31±0.01 ^c	0.31±0.00 ^c	0.26±0.01 ^b
	D	0.51 ±0.01 ^d	0.26±0.07 ^a	0.29±0.02 ^b	1.00±0.12 ^d	0.30±0.07 ^b
Zn	E	0.29 ±0.01 ^{bc}	0.44±0.04 ^c	0.52±0.07 ^d	1.00±0.12 ^c	0.38±0.12 ^b
	F	0.31 ±0.03 ^c	0.29±0.02 ^a	0.29±0.07 ^a	0.29±0.01 ^a	0.29±0.01 ^a
	G	3.02±0.01 ^e	0.43±0.00 ^c	0.32±0.02 ^c	0.32±0.02 ^c	0.28±0.00
	A	3.67 ±0.02 ^d	3.00±0.03 ^c	2.94±0.01 ^b	0.40±0.01 ^a	3.01±0.02 ^c
	B	2.97 ±0.01 ^d	2.25±0.43 ^b	2.39±0.00 ^c	0.54±0.01 ^a	3.02±0.07 ^c
	C	2.19 ±0.01 ^e	0.26±0.02 ^b	1.69±0.03 ^c	0.21±0.01 ^a	2.14±0.01 ^d
	D	2.95 ±0.02 ^e	2.79±0.37 ^c	2.81±0.02 ^{cd}	0.32±0.02 ^a	2.57±0.01 ^b
Pb	E	3.02 ±0.01 ^e	2.22±0.20 ^c	2.04±0.01 ^b	1.08±0.48 ^a	2.87±0.93 ^d
	F	2.45 ±0.01 ^c	3.00±1.01 ^d	3.02±0.02 ^{de}	0.30±0.01 ^a	2.02±0.02 ^b
	G	3.02 ±0.02 ^d	2.02±1.22 ^b	2.60±0.02 ^c	0.30±0.01 ^a	3.10±0.00 ^c
	A	0.22 ±0.05 ^{bc}	0.20±0.05 ^b	0.20±0.01 ^b	0.12±0.01 ^a	0.19±0.01 ^b
	B	0.19 ±0.00 ^a	0.18±0.02 ^a	0.23±0.02 ^b	0.34±0.00 ^c	0.22±0.07 ^b
	C	0.15 ±0.01 ^a	0.19±0.01 ^b	0.15±0.01 ^a	0.14±0.01 ^a	0.20±0.01 ^b
	D	0.28 ±0.01 ^e	0.20±0.03 ^b	0.22±0.01 ^{bc}	0.25±0.02 ^d	0.15±0.07 ^a
	E	0.21 ±0.00 ^a	0.20±0.02 ^a	0.24±0.07 ^b	0.31±0.04 ^d	0.28±0.06 ^c
	F	0.19 ±0.02 ^c	0.24±0.02 ^d	0.17±0.02 ^b	0.13±0.03 ^a	0.18±0.01 ^c
	G	0.22 ±0.00 ^d	0.23±0.01 ^d	0.18±0.02 ^c	0.10±0.00 ^a	0.16±0.02 ^b

Key: K- Potassium, Ca- Calcium, Cu- Copper, Fe- Iron Mn- Manganese, Pb- Lead, Zn- Zinc, A-100% fermented seeds of *P. africana*, B-90% fermented seeds of *P. africana* + 10% ginger, C-80% fermented seeds of *P. africana* + 20% ginger, D- 70% fermented seeds of *P. africana* + 30% ginger, E-90% fermented seeds of *P. africana* + 10% garlic, F-80% fermented seeds of *P. africana* + 20% garlic, G-70% fermented seeds of *P. africana* + 30% garlic

Note: mean in the same column with different superscripts are significantly different at ($p < 0.05$).

Conclusion and Recommendation

This study demonstrated that fortifying the fermented *P. africana* seeds with *Zingiber officinale* and *Allium sativum* enhanced its nutritional profile. Among the proportions, 70% fermented seeds of *P. africana* fortified with 30% ginger had highest antioxidant properties and vitamin C content, while the 90% fermented seeds of *P. africana* fortified with 10% garlic had highest fiber content. It suggests that ginger and garlic fortification may help boost the nutritional quality of fermented seeds of *P. africana*, providing a potential solution to address micronutrient deficiencies and also a promising alternative to seasoning salts and animal protein. This study suggests the need for further investigation to determine the shelf life and preservative effect of the fortified condiment. Furthermore, the use of these food enhancers could contribute to global efforts aimed at reducing food insecurity by improving the nutritional quality of local foods.

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References

- Achikanu C. E, Ani O. N & Akpata E. I. (2020). Proximate, vitamin and phytochemical composition of *Cucumis metuliferus* seed. *International Journal of Food Science and Nutrition*. 5(20), 20-24. ISSN: 2455-4898; Impact Factor: RJIF 5.14
- Agunwah I. M, Ogueke C. C, Nwosu J. N. & Anyogu A. (2024). Microbiological evaluation of the indigenous fermented condiment *okpeye* available at various retail markets in the south eastern region of Nigeria. *Heliyon*, 10(3). <https://doi.org/10.1016/j.heliyon.2024.e25493>
- Akpata E. I, Ani O. N, & Nwodo O. F. (2023). Phytonutrients and anti-nutrient composition of aqueous extract of fermented seeds of *Prosopis africana*. *Asian Journal of Research in Biochemistry*, 13(3), 12–27. <https://doi.org/10.9734/ajrb/2023/v13i3256>
- Akpi U. K, Nnamchi, C. I. & Ugwuanyi J. O (2020). Development of Starter Culture for the Production of African Condiments and Seasoning Agents. *Advances in Microbiology*. 10(12), 599–622. <https://doi.org/10.4236/aim.2020.1012044>
- Akpi, U.K., Zaini, N.A.M., Nnamchi, C.I., Ugwuanyi, J.O., Wan-Mohtar, W.A.A.Q.I & Noor, W.S.A.W.M. (2023). Biotechnology of *okpeye*: A Nigerian traditional fermented food condiment. *Advances in Microbiology*. 13, 373-385. <https://doi.org/10.4236/aim.2023.138024>
- Aremu M. O, Odoh R, & Bamidele T. (2015). Effect of processing on nutritional composition of African locust bean (*Parkia biglobosa*) and mesquite bean (*Prosopis africana*) seeds. *Communication in Applied Science*. 3(1), 22–41. SSN 2201-7372
- Asimi O. A, Sahu N. P & Pal A. K. (2013). Antioxidant activity and antimicrobial properties of some Indian spices. *International Journal of Science Research Publication*. 3(3),1-8. ISSN 2250-3153
- Balogun M. A & Oyeyiola G. P. (2012). Changes in the nutrient composition of *okpehe* during fermentation. *Pakistan Journal of Nutrition*. 11(3), 270–275. <https://doi.org/10.3923/pjn.2012.270.275>
- Balogun M. A, Oyeyiola, G. P, & Kolawole F. L. (2017). Comparative study of the physicochemical analysis of *Prosopis africana* seeds fermented with different starter cultures. *Croatian Journal of Food Science and Technology*. 9(1), 25–30. <https://doi.org/10.17508/CJFST.2017.9.1.04>
- Carazo A, Macáková K, Matoušová K, Krčmová L. K.; Protti, M.; Mladěnka, P (2021) Vitamin A update: forms, sources, kinetics, detection, function, deficiency, therapeutic use and toxicity. *Nutrients*. 13. 1703. <https://doi.org/10.3390/nu13051703>
- Egwin E, Musa A, Abubakar Y, & Mainuna B. (2013) Nigerian indigenous fermented foods: processes and prospects. *License Intech*. 7. <http://dx.doi.org/10.5772/52877>
- Eze V. C, Onwuakor C. E & Ukeke E. (2014). Proximate composition, biochemical and microbiological changes associated with fermenting African oil bean (*Pentaclethra macrophylla* Benth) Seeds. *American Journal of Microbiological Research*. 2(5), 138 - 142. <https://doi.org/10.12691/ajmr-2-5-3>
- Ezeikaibeya A. C, Nnenna, A. O & Kenechukwu, O. C. (2020). Proximate, phytochemical, and vitamin compositions of *Cucumis metuliferus* (Horned Melon) Rind. *Journal of Complementary and Alternative Medical Research*. 40–50. <http://doi.org/10.9734/jocamr/2020/v9i330144>
- Ezeocha C. V, Ugwuja J. L & Onyeabor, C. S. (2022). Evaluation of indigenous *okpeye* (*Prosopis africana*) processing conditions in Nsukka L.G.A, Enugu, Nigeria, and its effect on the quality of the fermented seasoning. *Nigerian Agricultural Journal*. 3(1),191-199.
- Fowoyo P. T. (2017). Microbiological and proximate analysis of *okpehe*, a locally fermented condiment. *Food & Nutrition Journal*. 5(6), 1–11. <https://doi.org/10.29011/2575-7091.100053>

- Ibeabuchi, J.C., Olawuni, I.A., Iheagwara, M.C., Ojukwu, M & Ofoedu, C. (2014). Microbiological evaluation of 'Iru' and 'Ogiri Isi' used as food condiments microbiological evaluation of 'Iru' and 'Ogiri Isi' used as food condiments. *Journal of Environmental Science, Toxicology and Food Technology*. 8(8), 45–50. <https://doi.org/10.9790/2402-08814550>
- Kumar K. N, Devadas S. M, Murugan S, Krishnan S. G, and Thayumanavan T (2018). Production and characterization of bacteriocin by lactic acid bacterium- *Pediococcus pentosaceus* NKSM1 isolated from fermented 'Appam' batter. *Journal of Pure and Applied Microbiology*. 12(3), 1315–1330. <http://dx.doi.org/10.22207/JPAM.12.3.34>
- Nwuche C. O. (2013). Isolation of bacteriocin-producing lactic acid bacteria from "Ugba" and "Okpiye", two locally fermented Nigerian food condiments. *Brazilian Archives of Biology and Technology*. 56(1), 101–106. <http://doi.org/10.1590/S1516-89132013000100013>
- Obidi O. F, Soyinka, O. O, & Kamoru T. A. (2023). Morphological, biochemical, and molecular characterization of bacteria isolated from water and submerged painted boat hulls in Badagry Lagoon, Lagos State, Nigeria. *Journal of Applied Science and Environmental Management* 27 (7) 1579-1589. <https://dx.doi.org/10.4314/jasem.v27i7.34>
- Ogbu J. C. (2024). Comparative proximate Analysis of *Citrullus colocynthis* (melon) and *Prosopis africana* (mesquite). *Research Square* 1–7. <https://doi.org/10.21203/rs.3.rs3824347/v1>
- Ojokoh, A. O & Eromosele O (2015). Effect of fermentation on proximate composition and microbiological changes of sorghum and pumpkin blend. *British Microbiology Research Journal*. 10(6), 1–14. Article no. BMRJ.19260. <https://doi.org/10.9734/BMRJ/2015/19260>
- Okafor U. I, Omemu A. M, Obadina A. O, Bankole M. O & Adeyeye S.A.O. (2018). Nutritional composition and antinutritional properties of maize ogi co-fermented with pigeon pea. *Food Science Nutrition*. 6, 424-439. <https://doi.org/10.1002/fsn3.57>
- Olajide M. A, Bello S. O & Lukman A. B. (2021). Minerals and vitamins composition of some fermented food condiments and complex salt-potassium sesquioxide. *International Journal of Research and Scientific Innovation*. 7(4). <https://dx.doi.org/10.51244/IJRSI.2021.8406>
- Olaniran A. F, Abiose S. H, & Hezekiah A. A. (2020). Garlic-ginger as a potential bio-preservative in fermented maize. *Journal of Microbiology, Biotechnology and Food Sciences*. 10(3), 467-473. <https://doi.org/10.15414/jmbfs.2020.10.3.467-473>
- Olaniran A. F & Abiose S. H (2019). Nutritional evaluation of enhanced un-sieved ogi paste with garlic and ginger. *Preventive nutrition and food science*, 24(3), 348–356. <https://doi.org/10.3746/pnf.2019.24.3.348>
- Ozioko, S. C., Madueke, A. C., & Anosike, C. A. (2020). Phytochemical composition and free radical scavenging activity of *Cucurbita maxima* fruit juice. *Research Journal in Advanced Sciences*, 1(1). <https://royalliteglobal.com/rjas/article/view/162>
- Popova, A & Mihaylova, D. (2019). Antinutrients in plant-based foods: A Review. *The Open Biotechnology Journal*. 13(1), 68–76. <https://doi.org/10.2174/1874070701913010068>
- Ryu J. H & Kang D. (2017). Physicochemical properties, biological activity, health benefits, and general limitations of aged black garlic: a review. *Molecules*. 22, 9-19. <https://doi.org/10.3390/molecules22060919>
- Ukaoma A, Nwachukwu M. O, Adjeroh L, Ezeanowai C, Iwu I & Duru M. C (2019). Comparative study on microbial loads of fermented castor oil seeds (*Ricinus communis*) and melon seeds (*Citrullus vulgaris*). *International Journal of Science and Research*. 8 (8). <https://doi.org/10.21275/ART20199530>
- Yeptho L & Ajungla, T. (2023). Proximate analysis and nutritional values of some fermented food products of Nagaland, India. *Journal of Psychopathological Research*, 61(1), 109-114. <https://doi.org/10.57023/JMycR.61.1.2023.109>