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Phytochemical, Antinutrient and Radical Scavenging Potential of *Parkia Biglobosa* Seeds as affected by Fermentation and Extraction methods

Evans Gmalen LOGTONG [1*] and Lydia QUANSAH [2]

[1] *Vaccines and Biological Products Department, Food and Drugs Authority, Cantonments, Accra, Ghana*
[2] *Department of Biotechnology, Faculty of Biosciences, University for Development Studies, Tamale, Ghana*

Correspondence: zoelogtong@gmail.com

ABSTRACT

Plants contain important bioactive compounds which possess radical scavenging ability. Processing methods such as fermentation and the kind of solvents used in extraction induces the plant to make available these bioactive compounds. Parkia biglobosa seeds are commonly fermented into a food spice called ''Dawadawa'' utilized as a condiment in soups and stews possessing varying nutritional and medicinal benefits. The study evaluated the phytochemical, antinutrients content and antioxidant potential of raw and fermented Parkia biglobosa seeds extracted either with methanol or water. Phytochemical presence in the seeds was influenced by extraction solvent and fermentation. Total Phenolic Content was found to be 30.05-147.52 and 21.59-141.41 mg GAE/g of extract in methanol raw and methanol fermented seeds, respectively. While the aqueous raw and fermented had 14.89-72.80 and 19.84-85.19 mg GAE/g, respectively. The fermented and raw Parkia biglobosa seed extracts exhibited great reducing and free radical scavenging properties by reducing ferric (Fe^{3+}) to ferrous (Fe^{2+}). Furthermore, fermentation reduced phytate and oxalate content in the unfermented seeds from 3.410 mg/g to 0.925 mg/g and 5.717 mg/g to 1.356 mg/g, respectively on the sixth day of fermentation. Thus, fermentation increases the phenolic content and the antiradical activity of Parkia biglobosa seed and enhances its nutritional value due to the reduction in the antinutrient content.

Keywords: *Parkia biglobosa*, seed, fermentation, antinutrient, phytochemical, antioxidant

INTRODUCTION

Parkia biglobosa is a deciduous perennial tree from the savannah zone of West Africa which bears fruit containing seed imbedded in a yellow pulp (Nyadanu, et al., 2017). The seeds are usually fermented into "Dawadawa" which is used as a condiment for spicing soups and stews in many West African countries (Odunfa, 1985). It is reported to be consumed in several communities in Ghana (Nyadanu et al., 2017). This plant has been shown to have nutritional benefits with medicinal importance and indigenous people utilizes the different parts of the plant to manage various ailments (Abioye et al., 2013) including those arising from oxidative stress.

Excessive oxidative stress causes damage to macromolecules and gives rise to several sicknesses such as cancer, atherosclerosis, diabetes, immunosuppression, nephrotoxicity, heart diseases and certain neurodegenerative disorders like Parkinson's disease, schizophrenia, Alzheimer's diseases, anxiety and depression (Andrade et al., 2018). Secondary bioactive metabolic compounds from medicinal plants like phenolic acids, flavonoids and diet-derived antioxidants like vitamin C have been shown to effectively remove free radicals by neutralizing their deleterious effect thereby preventing diseases due to such molecules (Sylvie et al., 2014). Natural antioxidants such as dietary flavonoids can repair a range of oxidative radical damage to cells and help impede the oxidation of biomolecules such as carbohydrates, lipids, proteins and DNA and aid in preventing lipid hydrolysis or oxidative rancidity in foods (Komolafe et al., 2017). However, the antioxidant activity of plant sources in the prevention or treatment of several conditions related to free radicals has received little attention coupled with the presence of antinutrients that hinder the plant's utilization (Elshama et al., 2018).

Antinutrients are bioactive compounds found in plant-based food which have the potential to interfere with nutrient absorption (Bello and Akinyele, 2007; Popova and Mihaylova, 2019). To remove these antinutrients from foods, different processing methods including fermentation are used.

Fermentation can enhance important secondary metabolites in food of plant origin. Furthermore, fermentation can ensure food security for millions of people across the world, especially the marginalized and vulnerable groups by reducing antinutritional factors in the food thereby making the food safe in addition to enhancing the nutrients availability in the diet (Hasan et al., 2014). When food containing antinutritional factors are consumed without any processing, it can lead to deleterious effects such as vomiting, bloating, headaches, nutritional deficiencies, rashes and many others (Popova and Mihaylova, 2019). Fermentation has been applied in the production of most commercial products including pharmaceuticals, foods, beverages and industrial enzymes (Marshall and Mejia, 2011; Hasan et al., 2014).

Thus, the study evaluated the bioactive metabolites, in-vitro antioxidant/antiradical properties and antinutrients composition of raw and fermented *Parkia biglobosa* seeds extracted using either methanol or water as the solvent.

MATERIALS AND METHODS

Plant material collection and processing

Parkia biglobosa seeds were sourced from an open market in Tamale, Northern region of Ghana and sorted to remove undesirable seeds from the lot. The raw seeds were washed under tap water and kept in aluminium pot and covered with enough water and brought to boil for 12 hr at 100°C. After boiling, the seeds were strained using metal strainer and opened-air dried and manually dehulled using mortar and pestle. Afterwards, 25 kg of the cooked and dehulled seeds were covered with enough water and boiled again at 100°C for 1 hr. The dehulled and cooked beans (initial pH 5.62) were divided into two batches with three replications of bulked seeds each for raw/control and fermented seeds. The raw seeds were kept in 4ºC while the seeds earmarked for fermentation were fermented spontaneously at an incubation temperature of 25ºC using a digital electronic biochemical incubator (Model number; DNP-9052A, Guangdong, China) with temperature range; room temperature +5 – 65°C. Sampling was done during the fermentation process every 24 hr for six days when the pH was 8.50. Samples (raw and fermented kept in 4ºC) were air-dried and homogenized using mortar and pestle and stored at 4ºC until further analysis. The raw and fermented *Parkia*

biglobosa seeds were used for the antinutrient analysis (day 1 and 2 samples were lost) while only the raw and day 6 fermented seeds were used for the phytochemical and antioxidant determination.

Seed extracts preparation

To prepare the extracts, 100 g of the homogenized seed sample was macerated in 1 L of either water or methanol in four 1L volumetric flasks and stored for 48 hr at room temperature in the pharmaceutical chemistry laboratory of the Department of Chemistry (University of Ghana, Legon, Accra, Ghana). The mixtures were filtered using Whatman® No.1 filter paper (4.25 cm) with the aid of a diaphragm vacuum pump and concentrated using a rotary evaporator within controlled temperatures (42ºC-50ºC and 80ºC-100ºC) for methanol and aqueous extracts respectively due to the different boiling points of the two solvents. The concentrated extracts were then evaluated immediately for phytochemicals present and the remaining extracts concentrated further and kept at 4°C until it was later needed for antioxidant evaluation.

Preparation of *Parkia biglobosa* seed extracts and ascorbic acid used as a standard

To prepare different concentrations of the samples for antioxidant analysis, the concentrations (50, 100, 150, 200, 250) mg/ml of each extract were prepared out of a stock solution through serial dilution for determining the total phenolic content and free radical scavenging activities of the extracts at the various concentrations, and the standardization curve using Ascorbic Acid as control which was prepared in the same procedure.

Determination of phytochemicals

The extracts were qualitatively screened to identify the presence or absence of some plant bioactive metabolites in the extracts. The specific phytochemicals or bioactive groups determined were flavonoids, phenols, reducing sugars, cardiac glycosides, terpenoids, alkaloids, coumarins and saponins using the standard procedures described by (Chintalapani et al., 2018).

To test for flavonoids, few drops of 20% NaOH solution was added to 2 ml of extract. The development of yellow color that changes to colorless with the addition of diluted hydrochloric acid indicated the presence of flavonoids.

For determination of phenol, 2 ml of each extract was taken and few drops of 5% aqueous $FeCl_3$ solution was added and observed for the formation of deep blue or black color which indicated the presence of phenols.

To determine reducing sugar, 5 ml of extracts was added to 5 ml of Fehling's solution and the mixture heated for about 2-3 min in a water bath at 70ºC, brick-red precipitate development in the test tube indicated a positive reaction.

For cardiac glycosides determination, 2 ml of glacial acetic acid was added to 5 ml of sample and then a few drops of 5% aqueous ferric chloride solution was added to the solution mixture. One (1) ml of concentrated sulfuric acid was cautiously added. The development of a brownish ring at the edge of the test tube indicates the presence of cardiac glycosides.

For Terpenoids determination, to every 2 ml of extract, 1 ml of chloroform was added, and drops of concentrated Sulfuric acid was added. Reddish-brown precipitate formation indicates the presence of terpenoids.

For determining alkaloid, 2 ml of extracts was pipetted into a test tube, and 5 drops of Wagner's reagent were added. The development of a reddish-brown precipitate or coloration shows the presence of alkaloids.

To determine coumarins, in two test tubes for each extract, 2 ml of the extract was pipetted and 0.5 mL of 10% NaOH was added into one of the test tubes for each extract in a heating water bath until boiling. The test tubes were cooled. Four (4) ml of distilled water was added to the other test tube (serving as control). If the liquid from the test tube with the NaOH solution is transparent or more transparent compared to the control test tube liquid (without NaOH), it shows a positive reaction. If HCl was added, and it loses its yellow color and form precipitates, then this indicates the presence of coumarins.

Saponins were determined by taken 2 ml of extract and adding 5 ml of water. The mixture was shaken vigorously, and observation made for the formation of a stable persistent frothing or foaming upon warming, which supports the presence of saponins.

Determination of *in vitro* antioxidant/antiradical activities of *P. biglobosa* seed extract

Determination of total phenolic content

Total Phenolic Content (TPC) of the seed extracts were determined using the Folin–Ciocalteau procedure (Ruto et al., 2018) with some modifications in volumes of solutions. Two (2) ml of each extract's concentrations (50, 100, 150, 200 and 250) mg/ml was added to 2.5 ml of 0.2 N Folin–Ciocalteau reagent and incubated for 5 min. Two (2) ml of 7.5 g in 100 ml of Na_2Co_3 was added and the total volume made up to 25 ml using distilled water. The solution was then reserved for incubation at room temperature for 2 hr. The absorbance was measured at 760 nm using a Cole-Parmer® UV spectrophotometer (United States) against the blank. Garlic acid was used to prepare a sequence of solutions that were used to acquire the standard calibration curve [$y = 0.0056x + 0.4827$, $R^2 = 0.9924$; where y = absorbance of extract, and X=concentration of extracts]. From the above regression equation, concentrations of extracts were calculated. The total phenolics contents were calculated from the formula: **C** =cV/m; where C = total phenolic content in mg GAE/g of extract, c = concentration (mg/ml) of extracts gotten from the calibration curve, V =volume of extract in ml and m = mass of extract in gram. The total phenolic content in all samples was expressed in milligram of Garlic acid equivalents (GAE) per gram of extract (Ruto et al., 2018).

Ferric reducing antioxidant power assay

Ferric reducing antioxidant power assay was determined using the method described by (Dluya et al., 2017) with little modification in volumes of solutions. Two (2) ml of each of the extract with concentrations (50, 100, 150, 200 and 250) mg/ml were mixed with 2.5 ml of 200 mmol/L sodium phosphate buffer (pH 6.6) and 2.5 ml of 1% potassium ferricyanide solution. The mixture was incubated at 50ºC for 2 mins and 2.5 ml of 10% trichloroacetic acid (w/v) was added and the mixture was centrifuged at 3000 rpm for 10 mins. Supernatants of 2.5 ml were mixed with 2.5 ml of deionized water and 0.5 ml of 0.1% of ferric chloride. Then 1.5 ml of the mixture was pipetted, and the absorbance was read at 700 nm using Cole-Parmer® UV spectrophotometer curvets (United States) against the blank. The reducing power of the extract was indicated by the increase in absorbance.

Nitric oxide radical scavenging activity

The procedure as described by (Sylvie et al., 2014) was used to determine the NO^- radical activity of the extracts. To 2.5 ml of each extract concentrations (50, 100, 150, 200 and 250) mg/ml, 2 ml of sodium nitroprusside solution with pH 7.4 was added. The solution was mixed and incubated or allowed to stand for 2½ hrs. One (1) ml of sulfanilic acid (0.33% in 20% of glacial acetic acid) was added to 0.5 ml of the sample mixture and incubated for additional 5 mins for aromatic amine to completely diazotize. Afterwards, 1

ml of Naphthyl ethylene diamine dihydrochloride (0.1%) was added and the sample mixture allowed to stand at room temperature for 25 mins. It was then read at 540 nm against the blank after a pink chromophore in diffused light was formed.

Determination of Hydroxyl (HO^-) radical activity

Hydroxyl radical scavenging activities of *P. biglobosa* seed extracts was determined using a method by (Sylvie et al., 2014) with modifications as suited for our plant materials. In summary, each of the extract concentration (50, 100, 150, 200, 250) mg/ml, 1.5 ml of each extracts were pipetted. Then, 0.6 ml of 1 mmol/L of $FeCl_2$, 0.9 ml of 1,10-phenanthroline (1 mmol/L), 2.4 ml of 0.2 mol/L phosphate buffer, pH 7.8 and 1.5 ml of sulfuric acid (0.17 mol/L) were added respectively. The sample mixtures were made uniform and incubated at room temperature for 5 mins. The absorbance was read at 560 nm in contrast to the blank. The percentages of FRAP, nitric and hydroxyl radical scavenging activity were calculated using the formula.

$$\%\ of\ inhibition = \frac{\text{Absorbance of the Ascorbic acid} - \text{Absorbance of test sample}}{\text{Absorbance of Ascorbic acid}}\ x\ 100$$

(Nnamdi et al., 2017).

The 50% inhibition concentration of extracts (IC 50%) was calculated and obtained using the inhibition curve by linear regression analysis. The results were expressed as mean ± SEM.

Determination of quantitative antinutrient content

The antinutrients; oxalate, phytate and cyanide content were determined using the procedure described by (Oluwaniyi and Bazambo, 2014).

The oxalate content was determined using 75 ml sulfuric acid of 3.0 M and added to 1 g of powdered sample, stirred and filtered. Afterwards, 25 ml of the filtered extract was titrated hot (80-90 °C) against 0.05 M $KMnO_4$ solution to the point when a faded pink color appeared that continued for at least 30 sec. The oxalate content was expressed in mg/g of sample.

For phytate content determination, 4 g of each sample was soaked in 100 ml of 2% HCl for 3 hrs and then filtered. Then, 25 ml filtrate was titrated against $FeCl_2$ standard solution using 0.3% ammonium thiocyanate as an indicator till a brownish yellow color appeared and vanished within 5 mins. Phytic acid was calculated according to (Onivogui et al., 2015).

For cyanide analysis, 4 g of each powdered sample was macerated in a beaker containing a solution of 40 ml of distilled H_2O and 2 ml of orthophosphoric acid. It was mixed, stoppered and left to incubate overnight at room temperature for all bounded hydrocyanic acid to be freed. Five (5) ml of the resultant mixture was distilled into 40 ml of distilled H_2O containing 0.1 g of NaOH pellets. The distillate was made up to 50 ml with distilled water and 20 ml of this was titrated against 0.01 M silver nitrate solution using 1.0 ml of 5% potassium iodide solution as the endpoint, specified by a faded but permanent turbidity.

Data analysis

Data from the antioxidant and antinutrient analyses were analyzed using Microsoft excel 2016 (Microsoft office 2016 suite).

RESULTS

Phytochemical presence in raw and fermented *Parkia biglobosa* seeds

All evaluated phytochemicals were present in the methanol fermented extracts while methanol raw, aqueous raw and fermented were deficient in some of the phytochemicals (Table 1).

Presence of total phenolic content

The results of Total Phenolic Content (TPC) of *P. biglobosa* seed extract indicated that the TPC varied in all the extracts. Methanol extract (raw and fermented) showed consistent increase with increase in extract concentration, but this trend was absent in the aqueous raw or fermented extract (Figure 1). Methanol extracts showed the highest TPC than the aqueous extracts (Figure 1)

TABLE 1**. Qualitative determination of phytochemical in raw and fermented *P. biglobosa* seed extracted with methanol and aqueous solution**

Phytochemical compound	Extraction and processing method			
	Methanol		Water	
	Raw	Fermented	Raw	Fermented
Flavonoids	+	+	+	+
Phenols	+	+	+	+
Reducing sugars	-	+	-	+
Cardiac glycosides	+	+	+	-
Terpenoids	+	+	-	-
Alkaloids	+	+	-	+
Coumarins	+	+	-	+
Saponins	+	+	+	+

KEY: + Present - Absent

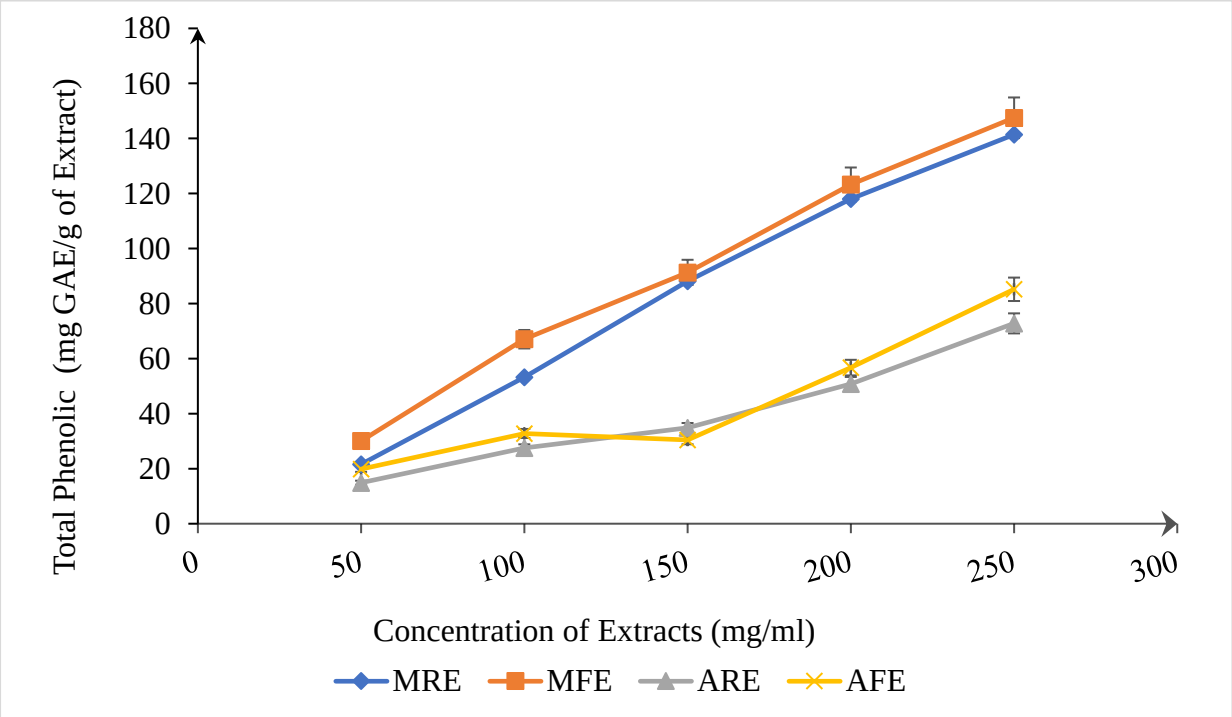

FIGURE 1. **Total phenolic content of raw and fermented *P. biglobosa* seed extracts.** MRE *(methanol raw extract)*, ARE (*aqueous raw extract), MFE (methanol fermented extract) and AFE (aqueous fermented extract).* Each point's *values are expressed as mean ± SEM, n =3 of bulked seeds.*

Ferric reducing antioxidant power (FRAP) assay

Methanol extracts reducing activity ranged from 12.3% - 54.9% for raw extracts and 17.7% - 63.1% for fermented extracts higher than the aqueous extracts with activity ranges of 8.3% - 32.4% for *aqueous raw extracts* and 14.3% - 28.2% for *aqueous fermented extracts* except for the control ascorbic acid with reducing activity range of 20.6% - 75.4% highest at all concentrations compared to all other extracts. Generally, the methanol extracts and the fermented seed extracts had higher reducing activity compared to the aqueous unfermented seed extract (Figure 2). In general, there was a dose dependent increase for all extracts.

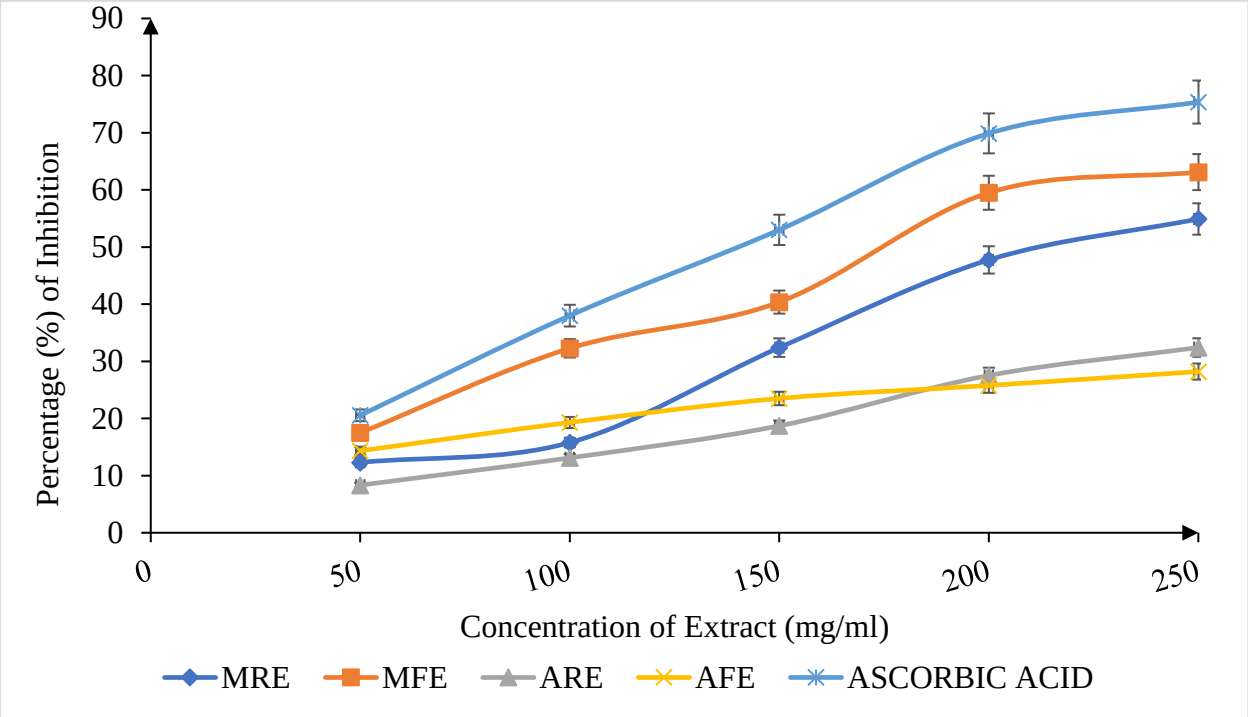

FIGURE 2. **Ferric reducing antioxidant power (FRAP) Activity of *P. biglobosa* seed extracts.** MRE *(methanol raw extract)*, ARE (*aqueous raw extract), MFE (methanol fermented extract) and AFE (aqueous fermented extract). Ascorbic acid was used as the control.* Each point's *values are expressed as mean ± SEM, n =3 of bulked seeds.*

Nitric Oxide Radical Scavenging Activity

The results of nitric radical activities of methanol and aqueous seed extracts of *P. biglobosa* seed expressed in inhibition percentages indicate that the aqueous raws and fermented seed extracts had the highest nitric radical scavenging activity of 66.1%-75.2% and 49.6%-69.4% compared to the methanol raw and fermented seed extract. All the seed extracts were able to scavenge and inhibit nitric radicals. However, the extract's radical inhibition % range was lower in comparison to ascorbic acid used as control (Figure 3). The percentage of inhibition values increased in all extracts and ascorbic acid with increasing concentrations (Figure 3)**.**

Hydroxyl radical scavenging activity

The results of hydroxyl radical activity of *P. biglobosa* seed extracts showed that all extracts exhibited scavenging activity on hydroxyl radicals (Figure 4).

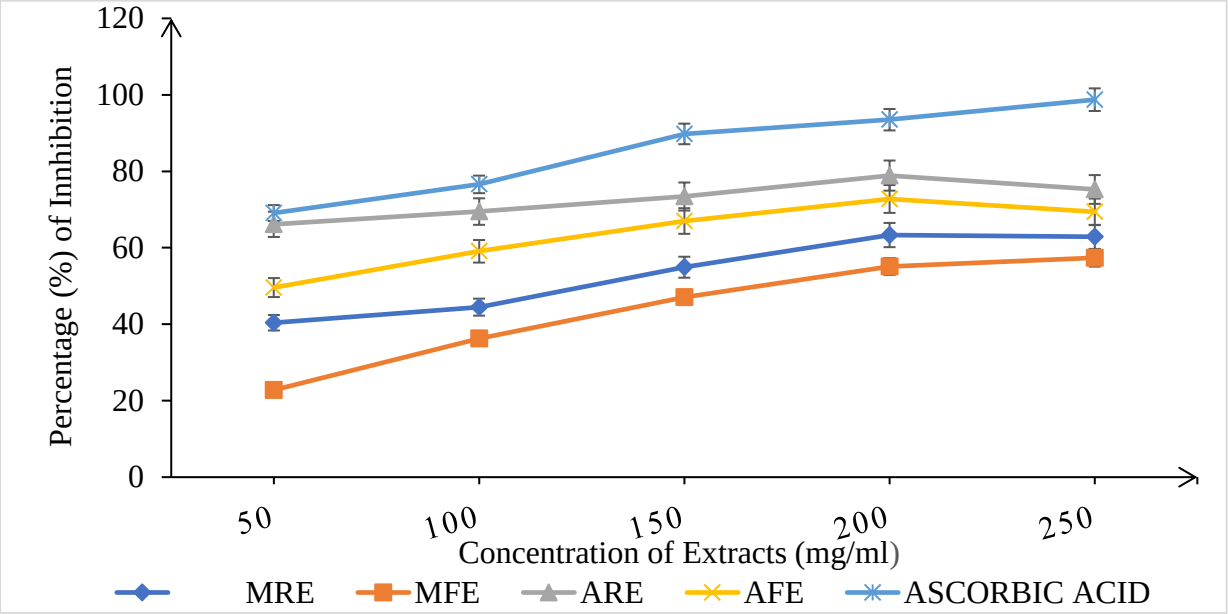

FIGURE 3. **Nitric Oxide radical scavenging activity of *P. biglobosa* seed extracts.** MRE *(methanol raw extract)*, ARE (*aqueous raw extract), MFE (methanol fermented extract) and AFE (aqueous fermented extract).* Each point's *values are expressed as mean ± SEM, n =3 of bulked seeds.*

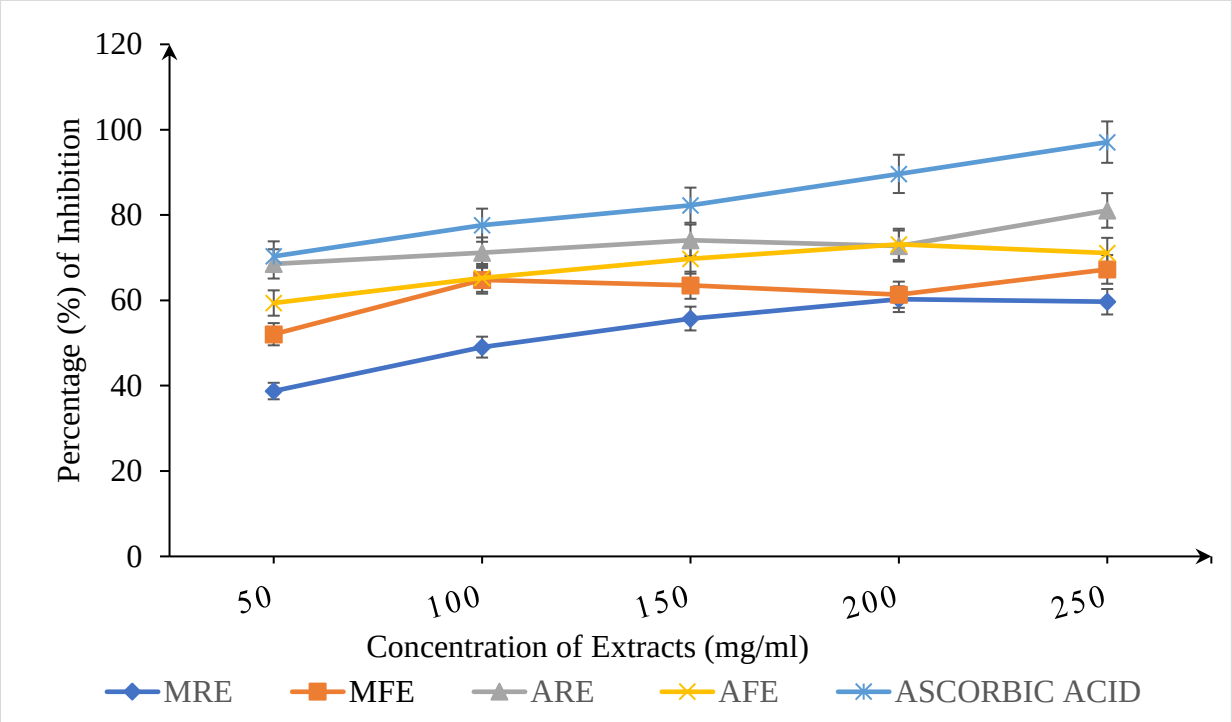

FIGURE 4**. Hydroxyl radical scavenging activity of raw and fermented *P. biglobosa* seed extracts.** MRE *(methanol raw extract)*, ARE (*aqueous raw extract), MFE (methanol fermented extract) and AFE (aqueous fermented extract).* Each point's *values are expressed as mean ± SEM, n =3 of bulked seeds.*

The $IC_{50\%}$ values of FRAP, NO^- and HO^- radical scavenging actions

The $IC_{50\%}$ (50% inhibitory concentration or half-maximal inhibition concentration) values of different radical scavenging tests (FRAP, NO^- and HO^-) of *P. biglobosa* seed were determined. FRAP showed the highest $IC_{50\%}$ compared to all the others while ascorbic acid had the lowest under each activity (Table 2**)**.

TABLE 2. **The minimum Inhibitory Concentration ($IC_{50\%}$) of raw and fermented *P. biglobosa* seed extracts on free radicals determined**

SAMPLE	FRAP		(NO^-)		(HO^-)	
	$IC_{50\%}$ (mg/mL)	R^2	$IC_{50\%}$ (mg/mL)	R^2	$IC_{50\%}$ (mg/mL)	R^2
MRE	255.456	0.978	150.213	0.791	153.830	0.740
MFE	181.553	0.977	185.190	0.922	117.767	0.566
ARE	383.894	0.993	80.995	0.557	78.129	0.551
AFE	430.136	0.865	112.759	0.693	96.917	0.583
AA	147.686	0.980	60.802	0.717	62.762	0.689

$IC_{50\%}$ (mg/mL) suitable to acquire 50% maximal scavenging activity at R^2 regression coefficient.

FRAP (Ferric reducing antioxidant power), NO^- (Nitric Oxide radical activity), HO^- (Hydroxyl radical activity).

Fermentation reduced the antinutrients composition of *Parkia biglobosa* seeds

The result of the antinutrients content analysis for phytate, oxalate and cyanide in the raw and fermented *P. biglobosa* seed samples revealed the presence of phytate and oxalate while cyanide was absent. The level of the antinutrients reduced as fermentation days progressed (Figure 5).

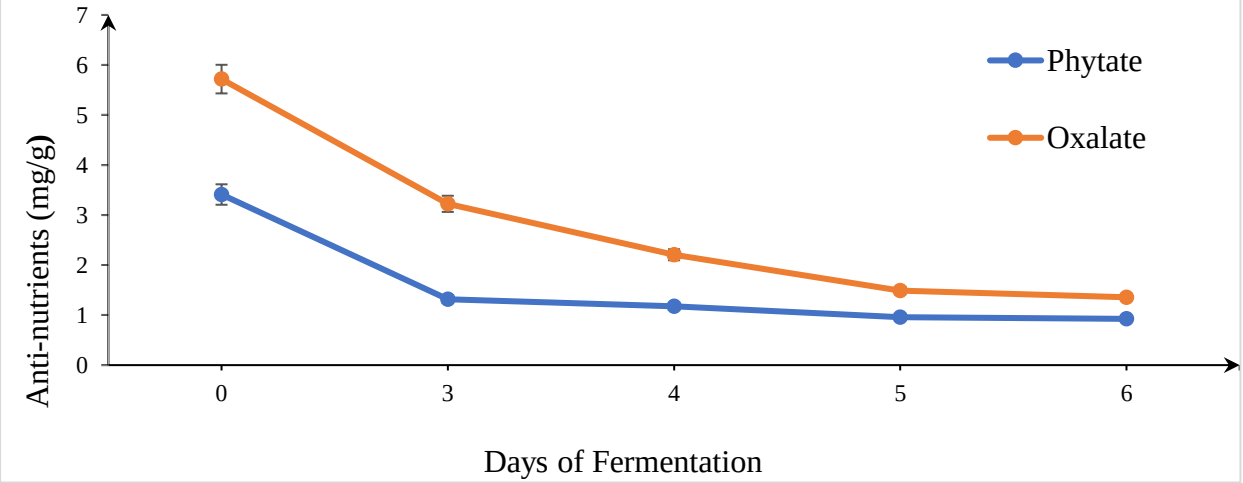

FIGURE 5. **Effect of fermentation on antinutrient composition in raw (day 0) and fermented (days 3-6) *P. biglobosa* seeds.** Each point's *values are expressed as mean ± SEM, n =3 of bulked seeds.*

DISCUSSION

The presence of more bioactive compounds in the methanol fermented samples point to the fact that fermentation and extraction with methanol enhances the release of phytochemicals in the solution including flavonoids and phenols and have an influence on the increase in total phenolic content of *Parkia biglobosa* seed. The abundance of the phenols (mg of GAE/g extract) and flavonoids concentration of both fermented and unfermented *Parkia biglobosa* seeds present a high potential for free radical scavenging (Singh and Sharma, 2018). Also, the reductive activity of the extracts is an indication of the presence of reductone antioxidants in the seed extracts which lead to the reduction or decrease of the ferric cyanide complex (Fe^{3+}) to ferrous cyanide (Fe^{2+}) form (Sylvie et al., 2014). There is an indication of the fermented seeds extracts having the ability to act on pro-oxidants in biological or food systems and help increase their shelf life (Hasan et al., 2014). The sequential removal of nitric oxides in solution by the extracts indicate that consumption of products made from fermented Parkia biglobosa seeds could help repair damages caused to cell macromolecules including Nucleic acids (DNA/RNA), proteins, carbohydrates and lipids (Singh and Sharma, 2018). The different *Parkia biglobosa* seed extracts evaluated indicate that the seed extract has an obvious nitric oxide radical scavenging effect in sodium nitroprusside in phosphate of buffer pH 7.4 and could be due to the presence of methanolic and water-soluble bioactive compounds with effective free radical scavenging properties like flavonoids and phenolics. The strong potential antioxidant action of the seed extracts could help eliminate the reported high reactivity of hydroxyl radical in the alteration in nitrogen bases of genes and inactivation of various proteins (Sylvie et al., 2014). These could also help reduce the already reported breakdown of amino acids such as phenylalanine into m-tyrosine and o-tyrosine by radicals, as this signalling molecules quickly cross cell membranes and react with every possible molecule in living organism especially with DNA, Proteins and Lipids and breakdown their functional chains leading to diseases (Sadananda et al., 2014; Singh and Sharma, 2018). This result corresponds with the report by Komolafe et al., (2017) which indicated that *P. biglobosa* extracts had free radical scavenging activity. Extracts are made of a mixture of several scavenging compounds which could act in a synergetic manner to enhance the antiradical activity.

Minimum inhibitory concentration or $IC_{50\%}$ is used to determine the minimum concentration that a compound scavenge for free radicals. The lower the $IC_{50\%}$ values, the higher the radical scavenging activity (Kaffoor et al., 2017). Thus, ascorbic acid showed higher scavenging activity compared with the seed extract for all concentrations. Notwithstanding, the seed extract showed good scavenging potential especially MFE for FRAP, ARE for NO^- and HO. Previous study by Ajaiyeoba, (2002) showed that extract from *Parkia biglobosa* seeds and leaves were able to inhibit growth of gram negative and gram positive bacteria though the standard gentamycin showed higher zone of inhibition compared to the leave and seed extracts.

The reduction in phytate and oxalate content with increasing days of fermentation show that the process of fermentation leads to a great reduction in the phytate and oxalate contents of the seeds. This indicates that fermentation possess the ability to biodegrade antinutrients in the *Parkia biglobosa* seeds thereby reducing its effects on nutrients bioavailability and absorption (Hasan et al., 2014). This suggest that fermented *Parkia biglobosa* seeds used for food has the

potential to mitigate the effect of specific nutrient deficiency especially in vulnerable populations in rural areas. Oxalates in plant samples could be in the form of soluble salts such as sodium, potassium or ammonium oxalate (Oluwaniyi and Bazambo, 2014). Fermentation is not only known to reduce antinutritional factors but also increase the quantities of amino acids such as aspartic acid, glutamic acid, valine and alanine which were analysed for in *Parkia biglobosa* seeds (Odunfa, 1985). Fermentation helps retain the nutrients that are usually destroyed during food processing such as cooking. These could mean that consuming fermented "Dawadawa" can enhance nutrient availability as it has less antinutritional factors.

CONCLUSION

This study established that *P. biglobosa* seeds extracted with methanol are rich in phytochemicals which could serve a potential source of bioactive compounds with pharmaceutical importance. It was also found that extract with methanol or aqueous solution enhances antioxidant properties and as the concentration of sample increased, the percentages of inhibition also increased which could indicate that *P. biglobosa* seeds have the potential to be used in the formulation of new antioxidant drugs that will prove beneficial in the treatment of free radical related disorders. Furthermore, the study revealed that fermentation reduced the amount of antinutritional factors (phytate and oxalate) present in the seeds which will enhance proper absorption of nutrients.

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