International Food Research Journal 26(6): 1707-1715 (December 2019)

Journal homepage: *http://www.ifrj.upm.edu.my*

Effect of natural fermentation on the physicochemical, nutritional, functional, and microbiological properties of baobab (*Adansonia digitata* L.) fruit pulp flour

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Article history

Received: 23 December, 2018
Received in revised form: 5 August, 2019
Accepted: 5 September, 2019

Keywords

Anti-nutrients
Baobab fruit pulp
Fermentation
Functional properties
Microbiological characteristics

Abstract

The changes occurring during the fermentation of baobab fruit pulp flour (BFPF), a raw material used to prepare a traditional dish in Sudan locally known as *kurundu*, were studied. The BFPF was naturally fermented at different durations (12, 24, and 36 h), dried and milled. The changes investigated included chemical composition, amino acid profile, anti-nutritional factors (phytic acid and tannin), mineral extractability, and microbiological, physicochemical and functional properties (bulk density, water and fat absorption capacity). Fermentation significantly increased the crude protein and fibre, functional properties and *in vitro* protein digestibility of BFPF throughout the fermentation period. Essential and non-essential amino acids were enhanced following fermentation with phenylalanine and aspartic acid, respectively, exhibiting the highest values after 24 h fermentation. Ascorbic acid and anti-nutritional factors significantly decreased throughout the fermentation period, with a concomitant increase in mineral extractability. Lactic acid bacterial, bacterial, and yeast and mould counts significantly increased after fermentation for 12 h with maximum value observed in BFPF fermented for 36 h. Total acidity increased with fermentation period, with a concomitant reduction of the pH. The improvement in the nutritional quality alongside with the reduction of the anti-nutritional factors during the fermentation of BFPF will contribute to recommending this process for wide application to increase the nutritional and health benefits of this important and staple fruit.

Introduction

Fruits from wild trees constitute a vital part of diets and sources of income among people in Africa. The products from these trees are usually eaten raw or consumed after traditional processing such as fermentation of the seeds and fruits, which enhances the nutritional, sensorial, and functional properties of such products (Parkouda *et al*., 2009). Baobab tree (*Adansonia digitata* L.) is one of the most remarkable trees that have been employed for centuries in many African countries to provide food, medicine and fodder (Gebauer *et al*., 2016). The pulp of baobab fruit is regarded as good source of vitamin C, pectin, and major minerals such as Ca, Mg and P (Kaboré *et al*., 2011). However, the fruit pulp of baobab has low nutritional composition in terms of protein, particularly essential amino acids, and fat contents (Osman, 2004; Abdalla *et al*., 2010). Moreover, baobab fruit pulp has naturally occurring anti-nutritional factors like phytates and tannins that can affect the bioavailability of macro- and micro-minerals in the fruits (Osman, 2004). In gastrointestinal tract, dietary minerals are chelated by these anti-nutritional factors thereby reducing their bio-availability and bio-accessibility (Idris *et al*., 2005). Exogenous and endogenous enzymes produced during domestic processing have been reported to significantly reduce the levels of anti-nutritional factors such as phytate and tannin of some fruits, cereals, and legumes. Reductions of such anti-nutritional factors, improvement of nutritional

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contents, microbiological characteristics, bioactive compounds, and antioxidant properties of baobab products obtained from different countries by processing methods such as fermentation, thermal treatment and acid or alkali treatment have been reported (Addy *et al*., 1995; Nnam and Obiakor, 2003; Parkouda *et al*., 2010; 2015; Tembo *et al*., 2017). Despite the fact that baobab fruits are used in various types of traditional foods and drinks in Sudan, internationally available information on the processing and utilisation of these incredible fruits is scarce (Gebauer *et al*., 2016). Consequently, reports on the effects of fermentation, a process used to prepare a traditional dish locally known as *kurundu*, on the nutritional and microbiological qualities of fermented fruits have not been documented yet. Therefore, the present work was conducted to evaluate the effect of fermentation on the chemical composition, amino acid profile, anti-nutritional factors, mineral contents and extractability, and microbiological, physicochemical and functional properties of baobab fruit pulp flour (BFPF).

Materials and methods

Sample collection and preparation

Baobab fruit was carefully cleaned to remove all foreign materials. The BFPF was prepared by using special high rate mechanical extracting machine to detach the seeds from the pulp and ground to pass through 0.4 mm mesh screen, and then stored. All chemicals and media used were of analytical grade. BFPF was traditionally fermented (lactic acid fermentation) as practiced by most Sudanese housewives (El Tinaysp *et al*., 1985). Natural fermentation was carried out by mixing the BFPF with water (1:7 w/v). The mixture was incubated at 37°C for different periods (0, 12, 24, and 36 h) in sterile covered flasks. This was followed by oven-drying (Heraeus UT 5042, Germany) of the fermented samples at 65°C for 16 h. Dried samples were ground to pass through a 0.4 mm screen and stored at 4°C in tightly closed containers until used in subsequent analyses.

Chemical composition

The amount of ash, crude protein, fibre, oil, and moisture of the raw and fermented BFPF were determined following the official method of analysis (AOAC, 2000). The sum of protein, oil, fibre, ash, and moisture contents was deducted from 100 to get the carbohydrate content.

Minerals composition

Mineral contents were determined following the Pearson method (Pearson, 1970) with some modifications. Briefly, 2 g of raw and fermented BFPF was ashed in a muffle furnace at 550°C for 3 h, and cooled. The ash obtained was treated with 10 mL concentrated hydrochloric acid (50% HCl) with addition of 5 mL of nitric acid (33%), and placed on water bath for 1 h. Then, 10 mL of HCl was added and placed on water bath again for 15 min, after that transferred to 100 mL volumetric flask filled to mark with distilled water, and then well shaken. After sample preparation, the element concentration was determined. Sodium (Na) and potassium (K) were determined by flame photometer. Calcium (Ca), magnesium (Mg), and iron (Fe) were determined by the atomic absorption spectrum. Phosphorus (P) was determined by the spectrophotometer (VV – visible spectrophotometer - wave length 440).

Hydrochloric acid extractability of minerals

The HCl extraction of minerals in the samples was determined following the method described by Chauhan and Mahjan (1988) with some modification. Briefly, sample (1 g) was added to 10 mL of 0.03 M HCl and then shaken for 3 h at 37°C. The mixture was then filtered, and the clear extract obtained was oven-dried at 100°C and then di-acid digested. The extractable minerals content was analysed as described above. The percentage of HCl extractability was calculated using Eq. 1:

$$\text{Mineral extractability (\%)} = \frac{\text{Mineral extractable in 0.03 M HCl (mg/100 g)}}{\text{Total minerals (mg/100 g)}} \times 100 \quad \text{(Eq. 1)}$$

Amino acid composition

The determination of amino acid contents was carried out following the official method (AOAC, 2000) with some modifications. Briefly, pulverised sample (500 mg) was placed in an evacuated sealed tube and hydrolysed with 6 N HCl (5 mL) at 100°C for 24 h. Before and after oxidation processes (H2O2/HCOOH, 24 h, chilled), the mixture was first adjusted to pH 2.2 with NaOH and filled with buffer (pH 2.2) to 100 mL mark before filtrating part of the mixture (2 mL) with membrane filter. The amino acids liberated were separated using Alpha Automatic Amino Acid Analyser (LKB Biochrom 4150) based on ion-exchange chromatography. The amino acids were detected at 570 nm except proline that was detected at 440 nm using a separate detector

channel. The amino acids were expressed as µg of amino acid per mg of protein.

Ascorbic acid content, pH, and titratable acidity determination

Ascorbic acid content was determined using 2,6-dicholoro-phenol indophenol dye reagent following the method described by Ruck (1963) with some modifications. Briefly, 100 mL of 0.4% oxalic acid and 30 mL of sample were blended for 2 min and then filtered through Whatman No. 1 filter paper. Oxalic acid of 0.4% was used to make up the filtrate to 250 mL. This was followed by titrating 20 mL of the filtrate against standard 2,6-dicholoro-phenol indophenol.

The pH was determined by thoroughly mixing 2% w/v of the flour sample in distilled water and the pH was measured with a pH meter (Hanna Instruments, Italy).

The total acidity was determined following the official method of analysis (AOAC, 2000). The total acidity was calculated using Eq. 2:

$$\text{Total acidity (mg lactic acid/100 g)} = \frac{T \times N \times EW \times 100}{\text{Sample weight} \times \text{Volume} \times 1000} \quad \text{(Eq. 2)}$$

where T = burette titre, N = normality of NaOH, DF = dilution factor (100 mL), EW = equivalent weight of lactic acid (90), weight of sample = 2 g, volume = 100 mL.

Microbiological evaluation

The total counts of bacteria (TCB), lactic acid bacteria (LAB), and yeast and mould (YM) of the raw and fermented BFPF samples were determined following the method described by Harrigan and McCance (1976) with slight modification. Briefly, the sample (1 g) was mixed with 9 mL of sterile peptone water (0.1%). Then, 1.0 mL of homogenate was diluted with 9.0 mL of 0.1% peptone water to prepare serial 10-fold dilutions. The TCB was determined by plating (pour-plate method) appropriate serial dilutions in duplicate on Plate Count agar and incubated at 37°C for 48 h. The LAB count was determined by plating appropriate serial dilution anaerobically on De Man, Rogosa and Sharpe agar and incubated at 37°C for 4 d. The YM count was determined by plating appropriate serial dilution on Malt Extract agar and incubated for 48 h at 28°C.

Functional properties

The method of Okezie and Bello (1988) was employed in the determination of bulk density. Water and fat absorption capacities were determined following the method described by Lin *et al*. (1974) with slight modification. Briefly, 3 g of the sample was centrifuged (Eppendorf, Germany) at 4,400 g for 30 min. Following incubation at room temperature, the increase in the sample weight was measured and results were expressed as percentage increase of the sample weight.

Anti-nutritional factors and in vitro protein digestibility

The determination of phytic acid of the samples was carried out following the method described by Wheeler and Ferrel (1971). Phytate phosphorous was calculated from standard curve, that was prepared and expressed as Fe(NO3)3 equivalent, by assuming iron to phosphorous molar ratio of 4:6. The vanillin-HCl method was used to determine the tannins content as described by Price *et al*. (1978). The IVPD was determined following the method described by Shastry and John (1991). A known sample weight containing 16 mg nitrogen was digested with 1 mg pepsin in 0.1 M HCl (15 mL) for 2 h at 37°C. Then, 15 mL of 10% trichloroacetic acid (TCA) was added to the mixture to stop the reaction, and it was then filtered through Whatman No. 1 filter paper. The nitrogen content in the filtrate was determined using micro-Kjeldahl method and the digestibility was calculated using Eq. 3:

$$\text{Protein digestibility (\%)} = \frac{\text{Nitrogen in filtrate}}{\text{Nitrogen in sample}} \times 100 \quad \text{(Eq. 3)}$$

Statistical analysis

Three fermentation batches were conducted, and all measurements were done in triplicates. The experiments were designed using a completely randomised block design and the effects of the treatments on the quality attributes of BFPF were statistically analysed using SAS/STAT software. The means was separated by Duncan multiple range test with a probability of $p \leq 0.05$.

Results and discussion

Effect of fermentation period on chemical composition of BFPF

The chemical composition of unfermented and fermented BFPF is shown in Table 1. The moisture content of unfermented sample (9.85%) significantly ($p \leq 0.05$) increased following fermentation, with

Table 1. Effect of fermentation periods on chemical composition of baobab fruit pulp flour.

Fermentation period (h)	Moisture (%)	Ash (%)	Protein (%)	Fat (%)	Fibre (%)	Carbohydrate (%)
0	9.85 ± 0.06^{d}	4.62 ± 0.05^{a}	4.28 ± 0.13^{c}	0.25 ± 0.01^{a}	4.05 ± 0.09^{c}	76.95 ± 0.05^{a}
12	10.17 ± 0.01^{c}	4.40 ± 0.00^{c}	4.41 ± 0.02^{c}	0.16 ± 0.03^{b}	4.26 ± 0.10^{c}	76.60 ± 0.04^{b}
24	10.68 ± 0.09^{a}	4.47 ± 0.02^{b}	4.81 ± 0.05^{b}	0.19 ± 0.02^{b}	4.70 ± 0.08^{b}	75.15 ± 0.09^{c}
36	10.26 ± 0.05^{b}	4.50 ± 0.03^{b}	5.01 ± 0.09^{a}	0.20 ± 0.00^{b}	5.20 ± 0.07^{a}	74.83 ± 0.03^{d}

Data are means of three determinations (n = 3) ± SD. Means with different superscripts in each column indicate significant differences at $p \leq 0.05$ based on Duncan multiple range test.

those fermented for 24 h having the highest value (10.68%). These values were higher than that of instant baobab fruit flour (7.78 - 8.59%) (Abdalla *et al*., 2010). However, the ash, fat, and carbohydrate contents of unfermented sample were significantly ($p \leq 0.05$) higher than that of fermented samples. In addition, the increase in fermentation period increased the ash and fat contents of the sample ($p \leq 0.05$). The ash content found in the present work is higher than 3.31% reported by Shukla *et al*. (2001). This contradicts the findings of higher ash contents following fermentation of pearl millet flour which was attributed to the utilisation of ash during microbial growth (Azhari *et al*., 2017). The result obtained in the present work showed that the raw sample had lower fat content as compared to that of baobab fruit (0.38%) reported by Shukla *et al*. (2001). The increase in fat contents throughout the fermentation period contradicts that observed in fermented pigeon pea (Adebowale and Maliki, 2011), and this could be due to the increase in the rate of lipolysis to fatty acids and glycerol during the fermentation process. The protein and fibre contents of the unfermented sample (4.28 and 4.05%, respectively) were significantly enhanced to 4.41 and 4.26%, respectively, following fermentation and this increased further throughout the fermentation period. These values are higher than that of baobab fruit (1.86 - 2.24%) reported by Muthai *et al*. (2017) but lower than that of baobab fruit pulp (5.15 - 5.75%) reported by Abdalla *et al*. (2010). Although the protein content of BFPF is lower than that in most legumes, it is greater than that in commonly consumed fruits like oranges (0.7%) and mangoes (0.6%) (Rathore, 2009). A similar trend of increase in protein content with fermentation period in baobab seed was reported by Addy *et al*. (1995). This increment in amount of protein during fermentation could be due to solubilisation of insoluble proteins of the flours and the synthesis of protein by fermenting microorganisms. The raw sample had crude fibre content lower than that reported by Osman (2004) for baobab fruit pulp flour (5.40%), but these increased throughout the fermentation period. According to Babalola and Giwa (2012), the decrease in crude fibre content occurring during fermentation could probably due to the degradation by fermenting microorganisms. However, this contradicts the findings in the present work.

Effect of fermentation period on mineral contents and extractability of BFPF

The result of mineral contents and extractability (Table 2) showed that K (547 mg/100 g), P (487.16 mg/100 g), and Mg (453.5 mg/100 g) were the major mineral elements of the unfermented BFPF, and from these, approximately 36.7, 35.5, and 68.22% of K, P, and Mg were extractable. The K content of raw baobab fruit obtained in the present work was lower

Table 2. Effect of fermentation periods on mineral contents and extractability of baobab fruit pulp flour.

Fermentation period (h)	Na	K	Ca	Mg	P	Fe
Mineral contents (mg/100 g)						
0	19.99 ± 0.03^{c}	547.00 ± 1.19^{d}	240.00 ± 2.73^{b}	453.50 ± 1.22^{d}	487.16 ± 2.07^{d}	5.01 ± 0.05^{d}
12	21.00 ± 0.09^{b}	570.00 ± 0.98^{c}	245.00 ± 2.66^{b}	465.00 ± 0.07^{c}	492.46 ± 1.13^{c}	5.27 ± 0.01^{c}
24	21.23 ± 0.12^{b}	636.00 ± 2.03^{b}	256.66 ± 1.00^{a}	474.40 ± 0.89^{b}	553.21 ± 2.75^{b}	5.30 ± 0.00^{b}
36	21.68 ± 0.08^{a}	665.00 ± 3.16^{a}	260.00 ± 3.45^{a}	500.00 ± 3.16^{a}	575.93 ± 5.01^{a}	5.83 ± 0.06^{a}
Mineral extractability (%)						
0	88.98 ± 0.05^{d}	36.66 ± 1.13^{d}	47.92 ± 0.07^{d}	68.22 ± 0.83^{d}	35.55 ± 1.99^{d}	33.42 ± 0.11^{d}
12	89.42 ± 0.13^{c}	42.85 ± 0.09^{c}	48.76 ± 0.00^{c}	71.66 ± 1.00^{c}	51.97 ± 1.15^{c}	44.97 ± 0.06^{c}
24	90.65 ± 0.08^{b}	52.72 ± 2.55^{b}	50.44 ± 0.91^{b}	77.06 ± 1.77^{b}	53.00 ± 0.04^{b}	55.24 ± 0.07^{b}
36	91.69 ± 0.11^{a}	64.04 ± 1.01^{a}	52.16 ± 0.88^{a}	80.32 ± 0.22^{a}	56.76 ± 0.95^{a}	56.59 ± 0.01^{a}

Data are means of three determinations (n = 3) ± SD. Means with different superscripts in each column indicate significant differences at $p \leq 0.05$ based on Duncan multiple range test.

than that of baobab fruit pulp (1,410 – 2,220 mg/100 g) from different countries as reported by Muthai *et al*. (2017). Na (19.99 mg/100 g) represented the least major mineral content with the highest extractability (88.98%). Trace mineral (Fe) content of the BFPF was 5.01 mg/100 g, and of this, about 33.42% was extractable. As shown in Table 2, the mineral contents were enhanced with fermentation time. However, fermentation of BFPF for 12 h had no significant effect on minerals of nutritional importance (Ca, P). Further increase in fermentation period significantly ($p \leq 0.05$) increased the contents of these minerals, with maximum values were obtained after fermentation for 36 h. During fermentation, the enzyme phytase may be released and then hydrolyses phytate, thereby resulting in the increase in the amount of P. Similarly, fermentation increased the mineral extractability of the BFPF and this significantly ($p \leq 0.05$) increased as fermentation period progressed. However, out of all the minerals analysed, Na was the most extractable (91.69%) while Ca the least (52.16%) after fermentation for 36 h. The increase in the HCl extractability of Na and Ca of high- and low-phytate corn genotypes after fermentation has been reported (Sokrab *et al*., 2014). In addition, the improvement in HCl extractability of Na and P has been observed following fermentation of malted millet, and this was attributed to lowering in the level of anti-nutrients (Abdelseed *et al*., 2011). The high HCl extractability of both major and trace minerals of BFPF may be due to hydrolysis and lowering of phytate and tannin by the enzymes phytase and tannase released during fermentation. The mechanism involves the removal of phosphate groups from the inositol ring of phytate, a process called dephosphorylation, which causes the decrease in the strength of mineral binding to the phytate, thereby enhancing the bioavailability of essential minerals (Sandberg *et al*., 1999).

Effect of fermentation period on amino acids profile of BFPF

Table 3 shows the effect of fermentation on amino acid profiles of BFPF. The essential amino acid composition of raw BFPF ranged between 6.4 and 22.0 mg/100 g, with histidine and isoleucine having the lowest and highest values, respectively. Fermentation for 12 h significantly ($p \leq 0.05$) increased both the essential and non-essential amino acids with lysine and phenylalanine being increased to 84.3 and 409.4 mg/100 g, respectively. However, histidine was not detectable after fermentation, and also non-essential amino acids such as aspartic acid and serine significantly ($p \leq 0.05$) decreased. The maximum values of essential and non-essential amino acids were from phenylalanine and aspartic acid, respectively, which were observed after 24

Table 3. Effect of fermentation periods on amino acid contents of baobab fruit pulp flour.

Amino acid	Fermentation period (h)				Recommended level (mg/100 g protein)*
	0	12	24	36	
Essential amino acids (mg/100 g)					
Histidine	6.4	n.d.	n.d.	n.d.	140
Isoleucine	22.0	202.3	189.6	175.6	400
Leucine	9.0	261.2	400.4	350.4	704
Lysine	7.0	84.3	122.1	161.4	544
Methionine	11.3	137.1	108.1	119.6	352
Phenylalanine	8.7	409.4	665.2	585.1	680
Threonine	15.5	63.5	90.9	92.1	400
Non-essential amino acids (mg/100 g)					
Aspartic acid	224.4	124.9	296.5	263.7	
Serine	68.3	55.4	49.5	47.6	
Glutamic acid	31.8	95.4	153.2	145.6	
Glycine	14.5	51.8	46.2	56.9	
Alanine	55.6	100.6	135.0	148.8	
Valine	15.9	137.3	194.0	178.0	
Cyst	16.1	238.5	n.d.	n.d.	
Tyrosine	8.7	261.6	431.3	392.9	
Arginine	20.4	135.6	162.2	203.4	
Proline	28.0	67.0	88.7	133.1	

Data are means of three determinations ($n = 3$). n.d. = not detected. *FAO/WHO/UN (1985).

h fermentation. The increase in amino acids after fermentation might be due to the metabolic activity of microorganisms involved in fermentation through which some amino acids might be utilised and others might be produced. Similarly, an increase in amino acids of baobab seeds following fermentation has also been reported (Parkouda *et al*., 2010). Regardless of the fermentation durations, all the essential amino acids in the samples were lower than those of recommended levels (FAO/WHO/UNU, 1985).

Effect of fermentation period on ascorbic acid, pH, total acidity, and microbial loads of BFPF

The effect of fermentation on ascorbic acid (AA), pH, and total acidity (TA) of BFPF is presented in Figure 1a. The AA content of unfermented BFPF was 263.52 mg/100 g, which is lower than that of instant baobab fruit (347.33 - 370.66 mg/100 g) reported by Abdalla *et al*. (2010). These differences could be attributed to environmental factors such as soil, climate, maturity, and season of the collection of the baobab fruits. It is apparent from the Figure that subjecting BFPF to fermentation process significantly ($p \leq 0.05$) lowered the AA content and this decreased further as fermentation progressed. Similarly, Oberman and Libudzisz (1985) reported that fermentation by lactic acid bacteria led to significant reduction in AA in fermented milk.

The pH value of BFPF was found to be 3.24 which is slightly higher than the pH values (3.03 - 3.20) obtained by Abdalla *et al*. (2010). The pH values decreased from 3.24 to 3.22, 3.20, and 3.18 as fermentation progressed from 0 to 12, 24, and 36 h, respectively. These results agree with those obtained by Nnam and Obiakor (2003) who reported that fermentation caused a rapid drop in pH of baobab seeds and rice grains.

The TA of BFPF was found to be 1.77 mg/100 g as shown in Figure 1a. Like in many traditionally fermented products, the TA increased with a concomitant decrease in the pH. In the present work, the TA increased from 1.77 to 1.79, 1.81, and 1.85 mg/100 g as fermentation progressed from 0 to 12, 24, and 36 h, respectively. Similar observation was

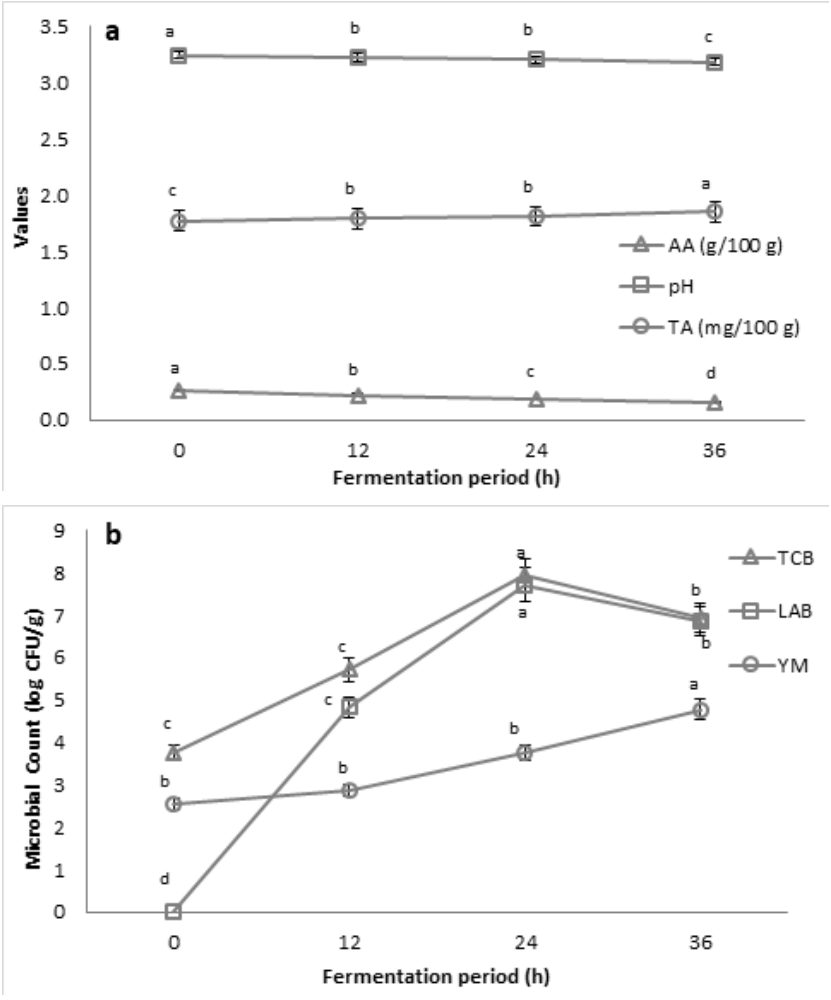

Figure 1. Effect of fermentation periods on ascorbic acid (AA), pH value, total acidity (TA) and microbiological characteristics of baobab fruit pulp flour. TCB: Total count of bacteria; LAB: Lactic acid bacteria; YM: Yeast and mould count; Error bars represent the standard deviation. Bars bearing different letters (a-d) are significantly different ($p \leq 0.05$) between fermentation periods.

reported by Nnam and Obiakor (2003). The decrease in pH with a concomitant increase in TA might be due to the activity of lactic acid bacteria.

The microbial loads of BFPF as affected by fermentation period are shown in Figure 1b. Fermentation for 12 h had no significant effect on the TCB and YM of BFPF as compared to unfermented BFPF. However, there was a sharp rise in LAB of BFPF after 12 h fermentation. In addition, as fermentation period progressed to 24 h, there was significant ($p \leq 0.05$) increase in TCB and YM of BFPF with a sharp decline in TCB after 36 h fermentation. Maximum YM was found in BFPF fermented for 36 h. In general, LAB were the dominant microflora and they remained dominant throughout the fermentation period. This explains that the natural fermentation of BFPF is generally lactic acid fermentation. This is similar to the findings reported by Parkouda *et al*. (2010) where LAB species (*Enterococcus faecium*) was the predominant microorganism during fermentation of baobab seed.

Effect of fermentation period on functional properties and anti-nutritional factors of BFPF

The effect of fermentation periods on functional properties of BFPF is shown in Figure 2a. The bulk density (BD) of unfermented sample (0.44 g/mL) increased following fermentation with maximum BD value was obtained in BFPF fermented for 36 h (1.00 g/mL). The BD values of the fermented samples were higher than that of fermented *Artocarpus altilis* pulp flour (0.46 g/mL) (Appiah *et al*., 2011). The increase in BD is needed because larger quantity may be packed within a constant volume which could offer greater packaging advantage for the fermented flour. Fermentation significantly ($p \leq 0.05$) decreased the fat absorption capacity (FAC) in raw BFPF from 1.76 to 1.29 mL/g. This result contradicts that reported by Appiah *et al*. (2011) where fermentation increased the FAC of breadfruit pulp flour. Further decrease in FAC was observed as fermentation progressed. The water absorption capacity (WAC) of unfermented sample (4.53 mL/g) significantly ($p \leq 0.05$) decreased after

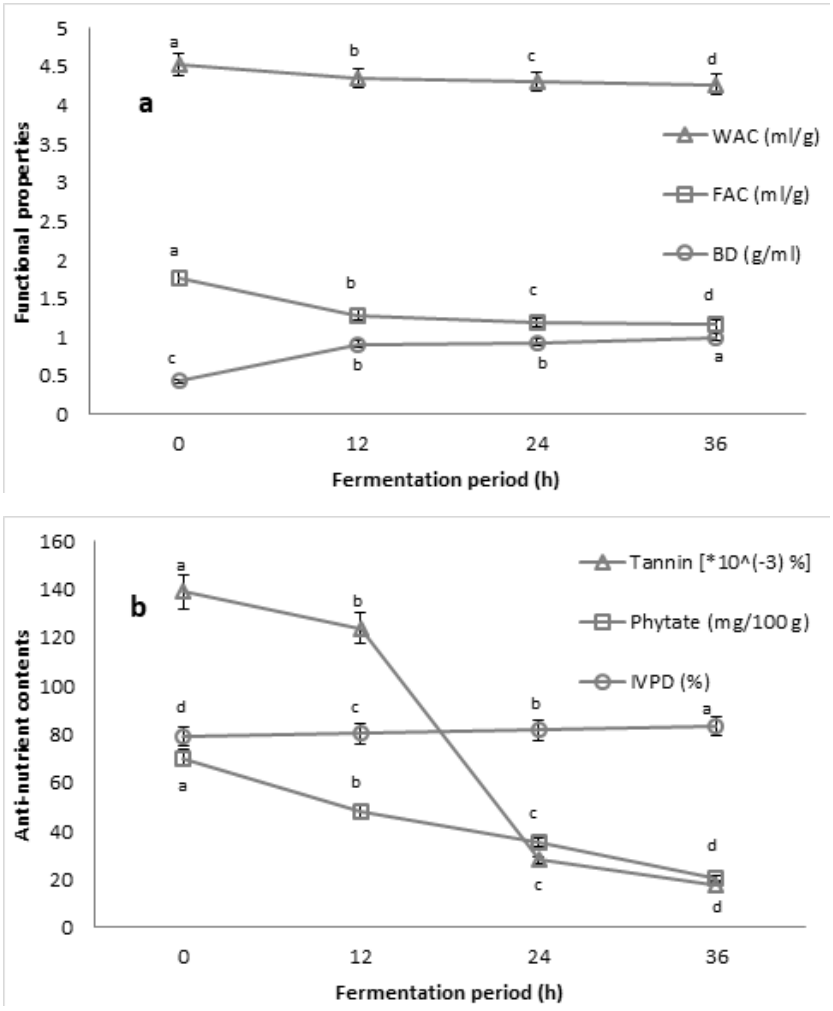

Figure 2. Effect of fermentation periods on functional and anti-nutrients of baobab fruit pulp flour. WAC: water absorption capacity; FAC: fat absorption capacity; BD: bulk density; IVPD: *in vitro* protein digestibility; Error bars represent the standard deviation. Bars bearing different letters (a-d) are significantly different ($p \leq 0.05$) between fermentation periods.

fermentation (4.35 mL/g). Similarly, Appiah *et al*. (2011) and Osungbade *et al*. (2016) have reported the decrease in WAC of *Artocarpus altilis* pulp flour and *Hura crepitan* seeds, respectively. As fermentation period progressed, there was further decrease in WAC of the samples.

The effect of fermentation period on anti-nutrient contents and IVPD of BFPF is presented in Figure 2b. Unfermented BFPF had 70.16 mg/100 g phytate and 0.139% tannin. These values were much lower than the phytate (2.6 mg/100 g) and tannin (0.9%) of BFPF in another study (Osman, 2004). The difference in the anti-nutritional factor contents could be attributed to the variations in the genetic makeup and environmental conditions. The IVPD of unfermented BFPF was found to be 79.31%, which is similar to that of fermented maize (Mohiedeen *et al*., 2010). Phytate and tannin contents significantly ($p \leq 0.05$) decreased after fermentation with a sharp decrease observed in tannin contents within the first 24 days. The decrease in phytic acid and tannin contents of the BFPF as fermentation progressed could probably be due to the action of degrading enzymes (phytase degrades phytic acid, tannase degrades tannin) produced by fermenting microorganisms, as also reported by other researchers (Idris *et al*., 2005; Abdelseed *et al*., 2011; Barthomeuf *et al*., 1994). However, fermentation significantly ($p \leq 0.05$) enhanced the IVPD of the sample with maximum IVPD being observed after 36 h of fermentation. The increase in IVPD values throughout the fermentation period is in agreement with other work (Mohiedeen *et al*., 2010), which could be attributed to partial degradation of complex storage proteins to simpler soluble products.

Conclusion

The present work demonstrated that natural fermentation of BFPF, which is usually carried out in traditional processing of this fruit, greatly reduced the anti-nutritional factors of the fermented product as compared to raw ones. In addition, fermentation could be a powerful method in improving the nutritional, total and extractable minerals and functional properties of BFPF. Fermentation of BFPF for 36 h is considered as the most suitable period to reduce the levels its anti-nutrient and enhance its nutritional composition, especially protein and fibre. Moreover, this process could be recommended for wide application to increase the nutritional and health benefits of this fruit and can also be utilised in the formulation of indigenous food products such as *kisra*.

References

Abdalla, A. A., Mohammed, M. A. and Mudawi, H. A. 2010. Production and quality assessment of instant baobab (*Adansonia digitata* L.). Advance Journal of Food Science and Technology 2(2): 125-133.

Abdelseed, B. H., Abdalla, A. H., Yagoub, A. E. G. A., Mohamed Ahmed, I. A. and Babiker, E. E. 2011. Some nutritional attributes of selected newly developed lines of sorghum (*Sorghum bicolor*) after fermentation. Journal of Agricultural Science and Technology 13(3): 399-409.

Addy, E. O., Salami, L. I., Igboeli, L. C. and Remawa, H. S. 1995. Effect of processing on nutrient composition and anti-nutritive substances of African locust bean (*Parkia filicoidea*) and baobab seed (*Adansonia digitata*). Plant Foods for Human Nutrition 48(2): 113-117.

Adebowale, O. J. and Maliki, K. 2011. Effect of fermentation period on the chemical composition and functional properties of pigeon pea (*Cajanus cajan*) seed flour. International Food Research Journal 18(4): 1329-1333.

Appiah, F., Oduro, I. and Ellis, W. O. 2011. Functional properties of *Artocarpus altilis* pulp flour as affected by fermentation. Agriculture Biology Journal of North America 2(5): 773-779.

Association of Official Analytical Chemists (AOAC). 2000. Official methods of analysis of Association of Official Analytical Chemists. 17th ed. United States: AOAC.

Azhari, A. M. N., Adiamo, O. Q., Awad, R. M. and Babiker, E. E. 2017. Changes in chemical composition and total energy as affected by fermentation and/or cooking of pearl millet flour supplemented with Moringa or fenugreek seeds flour. International Food Research Journal 24(4): 1562-1570.

Babalola, R. O. and Giwa, O. E. 2012. Effect of fermentation on nutritional and anti-nutritional properties of fermenting Soy beans and the antagonistic effect of the fermenting organism on selected pathogens. International Research Journal of Microbiology 3(10): 333-338.

Barthomeuf, C., Regerat, F. and Pourrat, H. 1994. Production purification and characterization of a tannase from *Aspergillus niger* LCF 8. Journal of Fermentation and Bioengineering 77(3): 320-323.

Chauhan, B. M. and Mahjan, H. 1988. Effect of natural fermentation on the extractability of minerals from pearl millet flour. Journal of Food Science 53: 1576-1577.

El Tinaysp, A. H., El Mahdi, Z. M. and El Soubki, A. 1985. Supplementation of fermented sorghum *kisra* bread with legume protein isolates. International Journal of Food Science and Technology 20(6): 679-687.

Food and Agriculture Organization of the United Nations (FAO), World Health Organization (WHO) and United Nations University (UNU). 1985. Energy and protein requirements: report of a Joint FAO/WHO/UNU

expert consultation. In World Health Organization Technical Report Series; no. 724. Geneva: WHO.

Gebauer, J., Adam Y. O., Sanchez, A. C., Darr, D., Eltahir, M. E. S, Fadl, K. E. M., ... and Kehlenbeck, K. 2016. Africa's wooden elephant: the baobab tree (*Adansonia digitata* L.) in Sudan and Kenya: a review. Genetic Resources and Crop Evolution 63(3): 377-399.

Harrigan, W. F. and McCance, M. F. 1976. Laboratory methods in food and dairy microbiology (revised edition). London: Academic Press.

Idris, W. H., AbdelRahman, S. M., ElMaki, H. B., Babiker, E. E. and El Tinay, A. H. 2005. Effect of germination, fermentation and cooking on phytic acid and tannin contents and HCl-extractability of minerals of sorghum (*Sorghum bicolor*) cultivars. Journal of Food Technology 3(3): 410-416.

Kaboré, D., Sawadogo-Lingani, H., Diawara, B., Compaoré, C. S., Dicko, M. H. and Jakobsen, M. 2011. A review of baobab (*Adansonia digitata*) products: effect of processing techniques, medicinal properties and uses. African Journal of Food Science 5(16): 833-844.

Lin, M. J. Y., Humbert, E. S. and Sosulski, F. W. 1974. Certain functional properties of sunflower meal products. Journal of Food Science 39(2): 363-370.

Mohiedeen, I. E., El Tinay, A. H., Elkhalifa, A. E. O., Babiker, E. E. and Mallasiy, L. O. 2010. Effect of fermentation on *in vitro* protein digestibility, protein fractions and amino acids composition of maize (*Zea mays* Linnaeus) cultivars. Electronic Journal of Environmental, Agricultural and Food Chemistry 9(5): 838-847.

Muthai, K. U., Karori, M. S, Muchugi, A., Indieka, A. S., Dembele, C., Mng'omba, S. and Jamnadass, R. 2017. Nutritional variation in baobab (*Adansonia digitata* L.) fruit pulp and seeds based on Africa geographical regions. Food Science and Nutrition 5(6): 1116-1129.

Nnam, N. M. and Obiakor, P. N. 2003. Effect of fermentation on the nutrient and antinutrient composition of baobab (*Adansonia digitata*) seeds and rice (*Oryza sativa*) grains. Ecology of Food and Nutrition 42(4-5): 265-277.

Oberman, H. and Libudzisz, Z. 1985. Fermented milks. In: Wood B. J. B. (ed). Microbiology of Fermented Foods, p. 308-350. United States: Springer.

Okezie, B. O. and Bello, A. B. 1988. Physicochemical and functional properties of winged bean flour and isolate compared with soy isolate. Journal of Food Science 53(2): 450-454.

Osman, M. A. 2004. Chemical and nutrient analysis of baobab (*Adansonia digitata*) fruit and seed protein solubility. Plant Foods for Human Nutrition 59(1): 29-33.

Osungbade, O. R., Gbadamosi, O. S. and Adiamo, O. Q. 2016. Effects of cooking and fermentation on the chemical composition, functional properties and protein digestibility of sandbox (*Hura crepitans*) seeds. Journal of Food Biochemistry 40(6): 754-765.

Parkouda, C., Ba/Hama, F., Ouattara/Songre, L., Tano-Debrah, K. and Diawara, B. 2015. Biochemical changes associated with the fermentation of baobab seeds in Maari: an alkaline fermented seeds condiment from western Africa. Journal of Ethnic Foods 2(2): 58-63.

Parkouda, C., Nielsen, D. S., Azokpota, P., Ouoba, L. I., Amoa-Awua, W. K., Thorsen, L., … and Jakobsen, M. 2009. The microbiology of alkaline-fermentation of indigenous seeds used as food condiments in Africa and Asia. Critical Reviews in Microbiology 35(2): 139-156.

Parkouda, C., Thorsen, L., Compaore, C. S., Nielsen, D. S., Tano-Debrah, K., Jensen, J. S., … and Jakobsen, M. 2010. Microorganisms associated with Maari, a baobab seed fermented product. International Journal of Food Microbiology 142(3): 292-301.

Pearson, D. 1970. The chemical analysis of foods. 6th ed. New York: Churchill Livingstone.

Price, M. L., Van Scoyoc, S. and Butler, L. G. 1978. A critical evaluation of the vanillin reactions as an assay for tannin in sorghum grain. Journal of Agricultural and Food Chemistry 26(5): 1214-1218.

Rathore, M. 2009. Nutrient content of important fruit trees from arid zone of Rajasthan. Journal of Horticulture and Forestry 1: 103-108.

Ruck, J. A. 1963. Chemical methods for analysis of fruits and vegetable products. Ottawa: Canada Department of Agriculture.

Sandberg, A. S., Brune, M. Carlsson, N. G. Hallberg, L. Skoglund, E. and Rossander-Hulthén, L. 1999. Inositol phosphates with different number of phosphate groups influence iron absorption in humans. American Journal of Clinical Nutrition 70(2): 240-246.

Shastry, M. and John, E. 1991. Biochemical changes and *in vitro* protein digestibility of the endosperm of germinating *Dolichos lablab*. Journal of the Science of Food and Agriculture 55(4): 529-538.

Shukla, Y. N., Dubey, S., Jain, S. P. and Kumar, S. 2001. Chemistry, biology and uses of *Adansonia digitata* – a review. Journal of Medicinal and Aromatic Plant Sciences 23: 429-434.

Sokrab, A. M., Mohamed Ahmed, I. A. and Babiker, E. E. 2014. Effect of fermentation on antinutrients, and total and extractable minerals of high and low phytate corn genotypes. Journal of Food Science and Technology 51(10): 2608-2615.

Tembo, D. T., Holmes, M. J. and Marshall, L. J. 2017. Effect of thermal treatment and storage on bioactive compounds, organic acids and antioxidant activity of baobab fruit (*Adansonia digitate*) pulp from Malawi. Journal of Food Composition and Analysis 58: 40-51.

Wheeler, E. L. and Ferrel, R. E. 1971. A method for phytic acid determination in wheat and wheat fractions. Cereal Chemistry 48(3): 312-320.