

Fermentation of Millet (*Pennisetum americanum*) and Pigeon Pea (*Cajanus cajan*) Seeds for Flour Production: Effects on Composition and Selected Functional Properties

J.C. Onweluzo and C.C. Nwabugwu

Department of Food Science and Technology, University of Nigeria, Nsukka, Nigeria

Abstract: The effects of period of fermentation on the chemical composition and selected functional properties of millet (*Pennisetum americanum*) and Pigeon pea (*Cajanus cajan*) seed flours were examined. The fermentation time ranged from 24-96 h. Flours of the unfermented seeds served as controls. Fermentation for 24 h decreased ($p < 0.05$) crude protein in both millet and pigeon pea flours. At 72 h, significant increase ($p < 0.05$) in crude protein occurred in pigeon pea. Ether extract and metabolizable energy increased ($p < 0.05$) in both flours at 72 h fermentation. Apparent decreases ($p > 0.05$) occurred in the total ash of both flours with increase in fermentation time except at the 96 h of pigeon pea. Tannin level was lowest in both flours at 72 h. Significant ($p \leq 0.05$) decreases in cyanide occurred in pigeon pea from the 24 h and was lowest at the 72 h. Phytate was low in both flours. A 10% (w/v) gruel of millet and pigeon pea flour exhibited 363 cp and 380 cp apparent viscosity respectively at 72 h fermentation. Significant ($p < 0.05$) reduction in water absorption capacity occurred only at the early stages (24 and 48 h) of fermentation in millet. Water Solubility Index increased in both flours with increase in fermentation period while reconstitution time reduced significantly ($p < 0.05$). Least gelation concentration increased by 100% in pigeon pea at 48 h and 72 h. Fermenting for 72 h seem to offer some advantages over other periods.

Key words: Fermentation, pigeon pea, millet, antinutrients, functional properties

INTRODUCTION

There is an increased interest in the production of flours from locally available and abundantly grown grains and pulses and fermentation is one of the choice methods employed. Fermentation is an age long method of processing cereals and legumes (Siegel and Fawcett, 1978). It modifies some physical characteristics of cereals and legumes, increases the level of some nutrients, digestibility and bioavailability (Hesseltine, 1965; WHO, 1998), decreases levels of antinutrients, increases nutrient density (Tomkins *et al.*, 1987; Nnam, 1999) and imparts some antimicrobial property (Mensah *et al.*, 1990; Mensah *et al.*, 1991). According to Quinn *et al.* (1975), fermentation of grains and oil seeds results in increased nutritional value and wholesomeness over the starting material and it may also lead to changes in vitamin levels. Fermentation actually holds promise as a food processing method that can be used to diversify the food uses of some under exploited plant foods like millet and pigeon pea.

Millet and Pigeon pea are good protein sources but they are underutilized. Millet (*Pennisetum americanum*) is one of the cereals produced extensively in Nigeria. Nigeria produces 21% of the world's total millet (FAO, 2002). Millet contains about 67% carbohydrate, and 12% protein. The seed is high in ash, iron, phosphorus and is an important source of the B group of vitamins (FAO, 1995). The essential amino acid profile of millet indicate that it contains more lysine, threonine, methionine and

cysteine than sorghum (FAO, 1995). Despite the rich nutrient content of millet, its use in Nigeria is limited to the production of household porridge-type breakfast gruel (akamu dawa) and dough (fura).

Pigeon pea (*Cajanus cajan*), a member of the leguminosae family has been reported to contain 20-22% protein, 1.2% fat, 65% carbohydrate and 3.8% ash (FAO, 1982). The mineral content and amino acid profile compares closely with those of soybean except in methionine content (Apata and Ologhobo, 1994; Osagie, 1998). Pigeon pea is not a very popular food in Nigeria, it is mainly resorted to during the hunger period (before harvest season). At such periods, mature dry pigeon pea is eaten in combination with starchy staples after several hours of boiling to soften the hard seed (Obizoba, 1983). Although pigeon pea and millet have potentials as good sources of much needed protein, they are underutilized in Nigeria (Eneche, 1999). Since emphasis has recently shifted from the use of special industrially produced weaning food and milk supplements to the use of simple household readily available staples processed by simple processing methods for use as a base for infant food formulation, a millet and pigeon pea blend could be a good complementary base for such formulation.

This study evaluates the effect of fermentation time on the composition and properties of millet and pigeon pea flours which could be used as a base for infant food formulation.

MATERIALS AND METHODS

Pigeon pea (*Cajanus cajan*) and millet (*Pennisetum americanum*) used for the study were procured from Nsukka market in Enugu State, Nigeria.

Preparation of materials: Two and half kilograms (2½ kg) of pigeon pea seeds were cleaned and divided into five equal portions of 500 g each. One portion was not fermented and it served as the control. The remaining four portions were separately steeped in deionized water in a ratio of 1:4 (w/v) (grain to water) and allowed to ferment by endogenous microflora at 28°C±2°C for 24, 48, 72 and 96 h respectively. At the end of each fermentation period, the seeds were separated from the steeping water by decanting. The samples were dried in an air oven at 55°C for 12 h. All the five portions were milled separately after drying in a laboratory hammermill to fine flour (1 mm mesh sieve). The samples were kept in screw capped jars until used for analysis.

Two and half kilograms (2½ kg) of millet seeds were cleaned and divided into five portions of 500 g each. One portion which served as the control was not fermented. The remaining four portions were steeped in water as described for the pigeon pea. At the end of each fermentation period the grains were separated from the steeping water dried and milled as described earlier. The flours were also kept in screw capped jars until used for analysis.

Chemical analyses

Proximate composition: Proximate composition of the flours were determined by standard methods of the AOAC (1990). Total nitrogen was determined by the micro-kjeldahl method. The crude protein was calculated by multiplying the total nitrogen (TN) by 6.25 conversion factor. Total ash was determined by incinerating at 600°C in an ashing muffle furnace. Crude ether extract was estimated by extracting with petroleum ether (40°C-60°C B Pt) using Tecator Soxtec apparatus. Nitrogen free extract was determined by difference. Energy was calculated by using the Artwater factor.

Anti-nutrients, cyanide, phytate and tannins determination

Cyanide: Cyanide was determined by the quantitative method described by Ikediobi *et al.*, (1980). Each flour sample (0.5 g) was extracted with 2 ml of 0.1 NHCl (pH 6.5). A known volume (0.5 ml) of the extract was incubated with 1.0ml of linamerase for 10 min at room temperature (28°C±2°C) and the volume was made up to 2 ml with 0.2 m sodium phosphate buffer (pH 6.8). A 5 ml volume of alkaline picrate was added to the mixture and incubation was continued in a water bath at 95°C for 5 min. The mixture was cooled to room temperature and the absorbance of the deep orange colour developed was read at 400 nm.

Phytate: Phytate was determined by a photometric method adopted from the method of Thompson and Erdman (1982). Each flour sample (2 g) was extracted with 100 ml of 2.4% HCl by shaking vigorously in a vortex mixer for 1 h at room temperature (28°C±2 °C). The suspension was filtered and 5 ml of the filtrate was mixed with 1 ml of 0.1 M Na-EDTA and 0.75 M NaOH in water made up to 25 ml with distilled water and placed on ion-exchange (AG1x4, 100-200 mesh) column. The column was washed with distilled water (15 ml) and 0.1 M NaCl (15 ml) before the sample was eluted with 15 ml of 0.7M NaCl. The eluate was collected and wet digested in a kjeldahl apparatus with a mixture of H₂SO₄ (0.5 ml) and HNO₃ (3 ml). The digest was cooled to room temperature, added 10 ml of distilled water and heated again for 10 min at 80°C. The solutions were transferred to 50 ml volumetric flasks, mixed with 2 ml of 2.5% ammonium molybdate solution in 1N H₂SO₄, 1 ml of sulphonic acid reagent and the volume was made up to 50 ml with distilled water. After mixing, each solution was allowed to stand for 15 min before the absorbance was read at 640 nm.

Tannin: Tannin content was determined by the vanillin-HCl method as described by Price and Butler (1977). Samples (0.6 g) were extracted for 60s at room temperature (28°C±2°C) with 3 ml of methanol. Extract was reacted with 3 ml of 0.1 M FeCl₃ in 0.1N HCl and 3 ml of 0.008M K₃Fe (CN)₆. Absorbance of the colour developed was read at 720 nm. Catechin was used as standard.

Functional properties

Apparent viscosity: Apparent viscosity was determined on the porridge (gruel) prepared with 10% (w/v) of each flour by heating at 100°C for 10 min. The determination was done with torsion viscometer according to the manufacturers instructions.

Water absorption capacity: Water absorption capacity was determined by a modification of the method of Lin *et al.* (1974). One gramme of each flour sample was weighed into a centrifuge tube and 10 ml of distilled water was added. Samples were vortexed for 5 min and allowed to stand for 15 min at room temperature (28°C ±2°C) before centrifuging (10,000xG) for 5 min. Excess water was decanted, the sample was allowed to drain by inverting the tube over absorbent paper. The weight, of water bound samples were determined by difference.

Water solubility index: Water solubility index was determined by the method of Onwulata *et al.* (1998). Two grams of each sample flour was weighed into porcelain dish and hydrated with 10 ml of distilled water. The hydrated flour was heated in a water bath at 100°C for 30 min and allowed to cool to room temperature. The

supernatant was decanted, weighed evaporated to dryness and weighed. Water solubility index was calculated as the weight percent of the dry supernatant.

Reconstitution time(s): Reconstitution time (in seconds) was determined by the method described by Nwanekezi *et al.* (2001). Two grams of each sample flour was spread on the surface of 50 ml of distilled water at room temperature ($28^{\circ}\text{C} \pm 1^{\circ}\text{C}$) in 150 ml cylinder. The time taken for the flour to completely disperse was recorded as the reconstitution time.

Data analysis: Determinations were done in 3 replicates. The Least Significant Difference test (LSD) was used to test differences between means. Statistical analysis was done by using the Genstat Release 4.24 DE (PC/Windows, 98) copyright 2003, Lawes Agricultural Trust (Rothamsted Experimental Station).

RESULTS AND DISCUSSION

Table 1 shows the proximate composition of the fermented and unfermented millet and pigeon pea flours. The crude protein values ranged from 8.90-9.46% and 20.15-22.04% for millet and pigeon pea flour respectively. The crude protein values of the unfermented flours agree closely with the values reported by Eneche (1999) for millet (11.3%) and by Onimawo and Osugo (2004) for pigeon pea (23.63). Millet showed 19% decrease in protein after 24 h fermentation and thereafter the protein increased ($p > 0.05$) to a maximum (9.5%) at the 72 h fermentation time. However the increase was not statistically significant ($p > 0.05$) when compared to the control. Pigeon pea exhibited 7 and 9% decreases in crude protein content at the 24 and 48 h fermentation period respectively. At 72 h fermentation period, pigeon pea had an apparent increase in crude protein. The decreases observed in crude protein during fermentation was attributed to a possible increase in the number of microorganism that use protein for metabolism. During fermentation, microorganisms hydrolyze proteins and it complexes to release free amino acids which can be used for the synthesis of new proteins (Frazier and Westhoff, 1978). This results agree with the report of Wang and Hesseltine (1981) and Ene-Obong and Obizoba (1996) who stated that fermentation process did not significantly change the total protein content and amino acid composition of substrates.

Fermentation increased crude ether extract in both millet and pigeon pea flour significantly ($p \leq 0.05$). The greatest increase in millet occurred at the 24 and 72 h while in pigeon pea, the increase from the 48 h were quite significant ($p \leq 0.05$). The increase were attributed to increased activities of lipolytic enzymes which hydrolyzed fat to glycerol and fatty acids. Similar observations were reported by Achinewhu and Isichei (1990), Obizoba and

Atii (1991) in fluted pumpkin and sorghum seeds respectively. Additionally release of some non lipid ether extractable materials released by fermenting micro-organisms may also have contributed to the observed increase in the crude ether extract of the samples. Increases in crude ether may have influenced the significant increase observed in metabolizable energy particularly at the 72 h of fermentation in both flours.

There were marginal decreases ($p > 0.05$) in total ash with increase in fermentation time in both millet and pigeon pea flours. The observed decrease in ash was attributed to possible losses of dry matter and volatiles which normally occur during fermentation (Quinn *et al.*, 1975; Nnam and Obiakor, 2003).

Fermentation significantly ($p \leq 0.05$) decreased total carbohydrate in millet within 24 h of fermentation and the decrease continued up to the 96 h when there was an apparent increase that did not actually differ statistically from the 24 h sample. In pigeon pea, the decreases observed were not statistically significant. The observed changes in carbohydrate with fermentation agrees with the report of Achinewhu and Isichei (1990) and Nnam (1995) on fermented fluted pumpkin seeds and fermented cowpea respectively. The apparent decreases were attributed to increased activity of amylolytic enzymes which hydrolyze starch and other complex carbohydrates to simpler sugars. The simpler sugars then provide energy for the fermenting micro-organism and carbon skeleton for possible synthesis of other compounds (Kazanas and Fields, 1981). Evidently such enzymic activities would also increase the availability of nutrients in fermented samples.

Anti-nutrients: The effects of fermentation on the tannin, phytate and cyanide levels in millet and pigeon pea flour are shown in Table 2.

The tannin levels ranged from 2.49-2.8 mg/g and 2.4-2.70 mg/g in millet and pigeon pea flour respectively. The tannin content of the unfermented pigeon pea used for the study was higher than the values (0.97-1.4 mg) reported by Ene-Obong (1995). The difference may be due to varietal differences. The polyphenols responsible for seed colour are predominantly located in the pericarp or testa and the darker the colour of the seed, the higher the levels of tannin (Deshpande and Cheryan, 1983). Millet showed about 8% decrease in tannin level within 24 h of fermentation and subsequently the tannin level decreased slightly ($p > 0.05$). The lowest tannin level was observed at 72 h fermentation.

Pigeon pea showed gradual marginal decreases ($p > 0.05$) in tannin level as fermentation progressed. The lowest tannin level was also observed at 72 h fermentation. The observed decreases in tannin with increase in fermentation time agrees closely with the report of Obizoba and Amaechi (1993) and Nnam (1995). The decreases were attributed to the hydrolysis

Table 1: Proximate composition of fermented and unfermented millet (*Pennisetum americanum*) and Pigeon pea (*Cajanus Cajan*) flours %

Sample	Moisture	Crude protein	Ether extract	Crude fibre	Total ash	Nitrogen free extract	Energy (Kcal)
UMF	6.8 ^d	9.18 ^d	1.5 ^e	3.0 ^a	3.7 ^a	75.82 ^a	353.5 ^c
FMF ₂₄	9.5 ^{ab}	7.45 ^c	3.7 ^b	2.9 ^a	3.2 ^a	73.25 ^b	356.1 ^c
FMF ₄₈	9.4 ^{ab}	9.20 ^d	3.1 ^c	2.8 ^a	3.5 ^a	72.01 ^c	352.74 ^{cd}
FMF ₇₂	8.6 ^c	9.46 ^d	4.5 ^a	3.0 ^a	2.9 ^{ab}	71.54 ^c	364.5 ^a
FMF ₉₆	8.1 ^c	8.90 ^d	3.5 ^b	3.0 ^a	2.7 ^b	73.80 ^b	362.3 ^a
UPF	9.7 ^{ab}	22.04 ^a	2.7 ^d	2.2 ^b	1.3 ^c	62.06 ^d	360.7 ^b
FPF ₂₄	10.2 ^a	20.40 ^c	2.8 ^d	2.1 ^b	1.0 ^c	63.50 ^d	360.8 ^b
FPF ₄₈	10.6 ^a	20.15 ^c	3.2 ^c	1.9 ^b	1.1 ^c	63.05 ^d	361.6 ^{ab}
FPF ₇₂	10.1 ^a	21.46 ^b	3.6 ^b	1.8 ^b	1.4 ^c	611.64 ^d	364.8 ^a
FPF ₉₆	9.4 ^b	20.61 ^c	3.5 ^b	2.1 ^b	2.3 ^b	62.09 ^d	362.3 ^a
LSD _(0.05)	0.67	0.56	0.24	0.40	0.64	1.002	2.9

Means with different superscript in the same column differ significantly ($p = 0.05$). $n = 3$

UMF	Unfermented millet flour	FMF ₂₄	Millet fermented for 24 h	FMF ₄₈	Millet fermented for 48 h
FMF ₇₂	Millet fermented for 72 h	FMF ₉₆	Millet fermented for 96 h	UPF	Unfermented pigeon pea flour
FPF ₂₄	Pigeon pea fermented for 24 h	FPF ₄₈	Pigeon pea fermented for 43 h	FPF ₇₂	Pigeon pea fermented for 72 h
FPF ₉₆	Pigeon pea fermented for 96 h				

Table 2: Effect of fermentation time on Tannin, Phytate and Cyanide content of Fermented and Unfermented Millet (*P. americanum*) and Pigeon pea (*Cajanus cajan*)

Samples	Tannin	Phytate (mg/g)	Cyanide (mg/g)
UMF	2.8 ^a	0.0117 ^b	5.1 ^b
FMF ₂₄	2.57 ^{ab}	0.0044 ^c	5.1 ^b
FMF ₄₈	2.56 ^{ab}	0.0028 ^c	4.9 ^b
FMF ₇₂	2.49 ^{ab}	0.0027 ^c	4.9 ^b
FMF ₉₆	2.60 ^a	0.0018 ^{cd}	4.9 ^b
UPF	2.70 ^a	0.1177 ^b	5.5 ^a
FPF ₂₄	2.60 ^a	0.0360 ^a	4.9 ^b
FPF ₄₈	2.47 ^{ab}	0.0323 ^a	4.7 ^{bc}
FPF ₇₂	2.45 ^{ab}	0.0240 ^b	4.5 ^c
FPF ₉₆	2.46 ^{ab}	0.0073 ^{bc}	4.5 ^c
LSD _(0.05)	0.32	0.0054	0.3

Means with different superscript in the same column differ significantly ($p = 0.05$) $n = 3$

UMF	Unfermented millet flour
FMF ₂₄	Millet fermented for 24 h
FMF ₄₈	Millet fermented for 48 h
FMF ₇₂	Millet fermented for 72 h
FMF ₉₆	Millet fermented for 96 h
UPF	Unfermented pigeon pea flour
FPF ₂₄	Pigeon pea fermented for 24 h
FPF ₄₈	Pigeon pea fermented for 43 h
FPF ₇₂	Pigeon pea fermented for 72 h
FPF ₉₆	Pigeon pea fermented for 96 h

of polyphenolic compounds or tannin complexes during fermentation. Obizoba and Atii (1991) reported that tannin-protein, tannic acid-starch and tannin-iron complexes are broken down during fermentation to release free nutrients. Since tannins are known to reduce the availability of proteins, carbohydrates and minerals through the formation of indigestible complexes, breakdown of such complexes will invariably improve the availability of the nutrients. This suggestion seem to be confirmed by Nnam (1999) who reported increased availability of proteins and minerals in children fed diets that had reduced tannin level through

fermentation. The 72 h fermentation time appear optimal for the production of low tannin flour from millet and pigeon pea.

Phytate: The levels of phytate were very low ranging from 0.002 mg-0.01 mg/g for millet and 0.02 mg-0.1 mg/g for pigeon pea. Egli *et al.* (2002) and Mulimani *et al.* (2003) reported higher values for millet (8.3 mg/g) and pigeon pea (184 mg-258 mg/g) respectively. Ene-Obong (1995) also reported a higher value (8.3 mg-11.3 mg/g) of phytate in pigeon pea than was obtained in the present study. However the disparity may be expected since according to Reddy *et al.* (1982) phytate levels vary in legumes and cereals with variety, cultivar type and soil type among other factors.

Significant decreases ($p \leq 0.05$) occurred in the phytate level of both millet and pigeon pea flour within 48 h of fermentation and subsequently the decreases became marginal ($p \geq 0.05$) up to the 72 and 96 h. The lowest level of phytate was attained at the 96h for both millet (0.0018 mg) and pigeon pea (0.0073 mg). The observed decreases in phytate with increase in fermentation time agrees closely with the report of Khetarpaul and Chauhan (1990) for fermented black gram. Mulimani *et al.* (2003) also reported a 1/3 reduction in the phytic acid content of soybean due to fermentation. The observed decrease in phytate levels was attributed to the activities of phytase during fermentation. According to Fardiaz and Markakis (1981), phytase dephosphorylates phytate on successive steps that terminates with the formation of inositol and phosphoric acid. The process is known to release certain metal like phosphorus thus increasing their availability.

Cyanide: The cyanide level ranged from 4.9 mg-5.1 mg/g and 4.5 mg-5.5 mg/g in millet and pigeon pea flours respectively. The cyanide levels observed in this study were low compared to values reported for some varieties of lima bean (210-3100 mg), sword bean and

Table 3: Effects of fermentation on selected functional properties of millet *P. americanum*) and Pigeon pea (*Cajanus cajan*) flours

Sample	Viscosity (cp)	Water absorption capacity (g/g)	Water solubility Index (%)	Reconstitution Time (s)	Least gelation concentration (%)
UMF	396.67 ^a	1.89 ^b	288.70 ^b	75 ^a	4.0 ^{cd}
FMF ₂₄	379.17 ^b	1.78 ^{bc}	325.9 ^a	69 ^b	4.0 ^{cd}
FMF ₄₈	375.0 ^b	1.68 ^{bc}	326.5 ^a	66 ^b	6.0 ^c
FMF ₇₂	363.33 ^c	1.65 ^{bc}	328.90 ^a	63 ^c	6.5 ^c
FMF ₉₆	356.67 ^c	1.50 ^c	331.90 ^a	57 ^d	6.5 ^c
UPF	389.64 ^a	2.19 ^a	226.55 ^d	66 ^b	5.0 ^c
FPF ₂₄	388.33 ^a	2.14 ^a	228.80 ^d	57 ^d	6.0 ^c
FPF ₄₈	381.67 ^b	2.04 ^a	269.40 ^c	54 ^d	8.0 ^b
FPF ₇₂	380.0 ^b	1.74 ^b	281.30 ^b	54 ^d	10.0 ^a
FPF ₉₆	375.0 ^b	1.42 ^c	281.50 ^b	53 ^d	10.0 ^a
LSD _(0.05)	8.46	0.26	9.34	1.2	

Means with different superscript in the same column differ significantly (p = 0.05) n = 3

UMF	Unfermented millet flour	FMF ₂₄	Millet fermented for 24 h	FMF ₄₈	Millet fermented for 48 h
FMF ₇₂	Millet fermented for 72 h	FMF ₉₆	Millet fermented for 96h	UPF	Unfermented pigeon pea flour
FPF ₂₄	Pigeon pea fermented for 24 h	FPF ₄₈	Pigeon pea fermented for 43 h	FPF ₇₂	Pigeon pea fermented for 72 h
FPF ₉₆	Pigeon pea fermented for 96 h				

jackbean (22 mg-46 mg/g) (Dela Vega and Sotelo, 1986) but higher than values reported for maize (0.12 mg/g), rice (0.4 mg/100 g), cowpea (0.61 mg/100 g) groundnut (0.79 mg/100 g) and papaya (1.4 mg/g) (Makinde and Lachance, 1989; Chandra *et al.*, 2005). However the observed levels of cyanide are within the safe limit. Significant ($p \leq 0.05$) decreases were observed in the cyanide level of pigeon pea during fermentation when compared with the control. In millet there were slight decreases in cyanide that were not statistically significant. The lowest cyanide levels were observed from the 72 and 48 h of fermentation in pigeon pea and millet respectively. Decreases in cyanide with increase in fermentation time has been associated with hydrolytic activities of enzymes from the fermenting microorganisms (Montgomery, 1980). Endogenous hydrolytic enzymes that occur in cyanophoric tissues may also have contributed to the hydrolysis of the cyanogens into non toxic sugar moiety and cyanohydrin.

Functional properties: Table 3 shows selected functional properties of the fermented and unfermented millet and pigeon pea flours.

Apparent Viscosity: Apparent viscosity of porridge (gruels) prepared with 10% ($\%v/v$) concentration of the flours ranged from 357-397 cp and 375-393 cp for millet and pigeon pea respectively. Fermentation decreased ($p < 0.05$) apparent viscosity in millet with in 24 h. Beyond 48 h the decreases were not statistically significant ($p > 0.05$). Similar decreases were also observed in pigeon pea from the 48 h. The decreases were attributed to the enzymic breakdown of higher molecular weight polysaccharide and polypeptides to lower molecular weight dextrins and peptides during fermentation. The implication of this decrease in viscosity is that gruels from the fermented flours would permit the addition of higher quantity of the solid without a concomitant increase in apparent viscosity. High nutrient density is a desirable characteristics in flours that can be used as a

base for infant food formulation. Nout *et al.* (1988) and Ariahu *et al.* (1999) had reported the viscosity reducing effect of fermentation.

Water absorption capacity: The Water Absorption Capacity (WAC) of the flours ranged from 1.50-1.89 g/g and 1.42-2.19 g/g for millet and pigeon pea flours respectively (Table 3). The WAC of the unfermented pigeon pea flour used in this study compares with the value (2.67%) reported by Onimawo and Osugo (2004) for raw pigeon pea. Water absorption capacity of the pigeon pea decreased significantly ($p \leq 0.05$) from the 72 h. In millet gradual decreases ($p > 0.05$) occurred in WAC as fermentation period increased. Apparently fermentation influenced the ability of the flours macromolecules to absorb water and this was probably reflected in the apparent viscosity exhibited by gruels from the flours. Elkhailifa *et al.* (2006) reported similar observation in fermented sorghum. Low water absorption is a desirable functional property required to produce thin gruels that can be used in infant formulas.

Water solubility index: Millet flours showed varied Water Solubility Index (WSI) from 288.7% for the unfermented to a range of 325.9-331.9% for the fermented samples (Table 3). Pigeon pea flours also showed similar increases in WSI with increase in fermentation time. The increases were expected since high molecular weight carbohydrates and proteins were hydrolyzed to simpler more soluble components during fermentation (Odufa, 1985; Achinewhu, 1986; Amadi *et al.*, 1999).

Reconstitution time: This property describes the ease of dispersibility of the flours. Millet flours showed higher ($p \leq 0.05$) reconstitution time that varied from 57-75s than the pigeon pea flour whose reconstitution time varied from 51-57s. Fermentation reduced reconstitution time significantly ($p \leq 0.05$) in both flours when compared with the controls. Fermentation may have induced a change

in the texture of the hydrophilic components of the flour to have influenced the ease of dispersibility of the flours. Ease of dispersibility is an important flour property in food formulation (Igene *et al.*, 2005).

Least gelation concentration: Millet flour showed a Least Gelation Concentration (LGC) range of 4.0-6.5% while in pigeon pea flour the LGC ranged from 5-10%. Significant increases ($p \leq 0.05$) occurred in both flours from the 48 h. In pigeon pea, the LGC increased ($p \leq 0.05$) at the 48 h and by the 72 h there was a 100 fold increase in LGC when compared to the control. Changes in LGC during fermentation was attributed to the enzymic hydrolysis of the carbohydrates and proteins. The implication of this observations is that since fermentation increased the flour concentration required for gel formation, blends of these fermented flours can be used in infant formula to enhance nutrient density. When flours that form gels at low concentration are used in infant formula, the diet would need a lot of dilution in an attempt to improve digestibility and this reduces the energy density in relation to volume (Ezeji and Ojmelukwe, 1993).

Conclusion: It was evident from the study that fermenting millet and pigeon pea for 24 h decreased crude protein content significantly ($p \leq 0.05$) beyond this period, fermentation had no significant change in the crude protein content of millet. Fermentation increased ($p \leq 0.05$) crude ether extract and reduced ($p \leq 0.05$) the levels of tannin, phytate and cyanide in both flours. Fermented millet and pigeon pea flours have improved functional characteristics that make them good base ingredients in infant food formulation. Fermenting for 72 h offer some advantages over other periods evaluated.

REFERENCES

- Achinewhu, S.C., 1986. Some biochemical and nutritional changes during the fermentation of fluted pumpkin (*Telferia occidentalis*). *Qualitars Plantarum: Plant Foods for Human Nutr.*, 36: 97-107.
- Apata, D.F. and A.D. Ologhobo, 1994. Biochemical evaluation of some Nigerian legume seeds. *Food Chem.*, 49: 333-338.
- A.O.A.C., 1990. *Official Methods of Analysis*. Washington DC: Association of Official Analytical Chemists.
- Achinewhu, S.C. and M.O. Isichei, 1990. The nutritional evaluation of fermented fluted pumpkin seeds (*Telferia occidentalis* Hook). *Discovery and Innovation*, 2: 62-65.
- Amadi, E.N., I.S. Barimalaa and J. Omosigbo, 1999. Influence of temperature on the fermentation of bambara groundnut (*Vigna subterranean*) to produce a dawadawa-type product. *Plant Foods for Human Nutr.*, 54: 13-20.
- Ariahu, C.C., U. Ukpabi and K.O. Mbajunwa, 1999. Production of African breadfruit (*Treculia Africana*) and Soybean (*Glycine max*) seed based food formulations, 2: Effects of germination and fermentation on microbiological and physical properties. *Plant Foods for Human Nutr.*, 54: 207-216.
- Chandra, A.K., D. Lahari and S. Mukhopadhyay, 2005. Goitrogen content of cyanogenic plant food of Indian origin. *J. Food Sci. and Technol.*, 42: 95-98.
- Deshpande, S.S. and M. Cheryan, 1983. Changes in phytic acid tannins and trypsin inhibitors activity on soaking of dry beans (*Phaseolus vulgaris* L.). *Nutrition Report International*, 27: 317-377.
- Dela Vega, A. and A. Sotelo, 1986. The nutritional quality and toxin content of wild and cultivated lima beans (*Phaseolus lunatus*). *Qual Plant. Plant Foods for Human Nutr.*, 36: 75-83.
- Ezeji, C. and P.C. Ojmelukwe, 1993. Effect of fermentation on the nutritional quality and functional properties of infant food formulations prepared from bambara-groundnut, fluted pumpkin and millet seeds. *Plant Foods for Human Nutr.*, 44: 267-276.
- Ene-Obong, H.N., 1995. Content of anti-nutrient and in vitro protein digestibility of the African yambean, pigeon pea and cowpea. *Plant Foods for Human Nutr.*, 48: 225 -233.
- Ene-Obong, H.N. and I.C. Obizoba, 1996. Effect of domestic processing on the cooking time, nutrients, antinutrients and in vitro protein digestibility of the African yeambean (*Sphenostylis stenocarpa*). *Plant Foods for Human Nutr.*, 49: 43-52.
- Eneche, E.H., 1999. Biscuit making potentials of millet/pigeon pea flour blends. *Plant Foods for Human Nutr.*, 54: 21-27.
- Egli, I., L. Davidsson, M.A. Tuillert, D. Barclay and R.F. Hurrell, 2002. The influence of soaking and germination on the phytase activity and phytic acid content of grains and seeds potentially useful for complementary feeding. *J. Food Sci.*, 67: 3484-3488.
- Elkhalifa, A.E.O., B. Schiffler and R. Bernhardt, 2006. Some physio-chemical properties of fermented sorghum flour. *J. Food Sci. and Technol.*, 43: 26-27.
- Frazier, W.C. and D.C. Westhoff, 1978. *Food Microbiology*. McGraw Hill Publishing Co. Ltd New York.
- Fardiaz, D. and P. Markakis, 1981. Degradation of phytic acid in oncom (fermented peanut press cake). *J. Food Sci.*, 46: 523-525.
- FAO, 1982. *Legumes in Human Nutrition Food and Agriculture Organisation of the United Nations, Food and Nutrition Series No. 2*, Rome.
- FAO, 1995. *Sorghum and Millets in Human Nutrition Food and Agriculture of the United Nations, Food and Nutrition Series No. 27*, Rome.
- FAO, 2002. *FAO production year book, 2001 vol. 55. Food and Agriculture Organisation of the United Nations Statistics Series No. 106*, Rome.

- Hesseltine, C.W., 1965. A millennium of fungi, food and fermentation. *Mycologia*, 62: 149-197.
- Ikediodi, C.O., G.O.C. Onyia and C.E. Eluwah, 1980. A rapid and inexpensive enzymatic assay for total cyanide determination in cassava (*Manihot esculenta* Crantz) and cassava products. *J. Agric. and Biol. Chem.* 4: 2803 -2809.
- Igene, F.U., S.O. Oboh and V.A. Aletor, 2005. Effects of some processing techniques on the functional properties of winged bean seed flours. *J. Food, Agric. and Environ.*, 3: 28-31.
- Kazanas, N. and M.L. Fields, 1981. Nutritional improvement of sorghum by fermentation. *J. Food Sci.*, 49: 819-821.
- Khetarpaul, N. and B.M. Chauhan, 1990. Fermentation of pearl millet flour with yeasts and lactobacilli: In vitro digestibility and utilization of fermented flour for weaning mixtures. *Plant Foods for Human Nutr.*, 40: 167-173.
- Lin, M.J.Y., E.S. Humbert and F.W. Sosulski, 1974. Certain functional properties of sunflower meal products. *J. Food Sci.*, 39: 368-370.
- Montgomery, R.D., 1980. Cyanogens in I. E. Liener (Eds.) *Toxic constituents of plant food stuffs*. Academic Press, New York, pp: 143-157.
- Makinde, M.A. and P.A. Lachance, 1989. Optimization of protein nutritive value of Ogi. *Nigerian J. Nutr. Sci.*, 10: 85 -93.
- Mensah, P.P.A., A.M. Tomkins, B.S. Draser and T.J. Harrison, 1990. Fermentation of cereals for reduction of bacterial contamination of weaning foods in Ghana. *Lancet*, 336: 140-143.
- Mensah, P.P.A., A.M. Tomkins, B.S. Draser and T.J. Harrison, 1991. Antimicrobial effect of fermented Ghanain maize – dough. *J. Appl. Bacteriology*, 70: 203- 210.
- Mulimani, V.H., N.S. Kadi and S. Thippeswamy, 2003. Effect of processing on phytic acid content in different red gram (*Cajanus cajan* L.) varieties. *J. Food Sci. and Technol.*, 40: 371-373.
- Nout, M.J.R., J.G.A.J. Hautvast, F.V.D. Haar, W.E.W. Marks and F.M. Rombouts, 1988. Formulation and microbiological safety of cereal-based weaning foods. In: D. Alwick, S. Moses and O. G. Schmidt (Eds.) *Improving young child feeding in Eastern and Southern Africa: Household-level Food Technology*, Ottawa International Development Research Centre, pp: 245- 260.
- Nnam, N.M., 1995. Evaluation of nutritional quality of fermented cowpea (*Vigna unguiculato*) flours. *Ecology of Food and Nutr.*, 33: 273-279.
- Nnam, N.M., 1999. Chemical and organoleptic evaluation of complementary foods formulated from maize, cowpea and plantain flours. *Nigerian J. Nutr. Sci.*, 20: 35-40.
- Nnam, N.M. and P.N. Obiakor, 2003. Effect of fermentation on the nutrient and antinutrient composition of Baobab (*Adansonia digitata*) seeds and rice (*Oryza sativa*) grains. *Ecology of Food and Nutr.*, 42: 265-27.
- Nwanekezi, E.C., N.C. Ohaji and O.C. Afam-Anene, 2001. Nutritional and organoleptic quality of infant food formulation from natural and solid state fermented tubers (cassava, sprouted and unsprouted yam)- soybean flour bland. *Nigerian Food J.*, 19: 55-62.
- Obizoba, I.C., 1983. Growth and nitrogen utilization of rats fed pigeon pea- rice mixtures. *Nigerian J. Nutr. Sci.*, 4: 35-40.
- Odunfa, S.A., 1985. Microbiological and toxicological aspects of fermentation of castor oil seeds for Ogiri Production. *J. Food Sci.*, 50: 1758-1759.
- Obizoba, I.C. and J.V. Atii, 1991. Effects of soaking, sprouting, fermentation and cooking on nutrient composition and some anti-nutritional factors of sorghum (guinea) seeds. *Plant Foods for Human Nutr.*, 41: 203-212.
- Obizoba, I.C. and N.A. Amaechi, 1993. The effect of processing methods on the chemical composition of baobab (*Adansonia digitata* L.) pulp and seed. *Ecology of Food and Nutr.*, 29: 199-204.
- Onwulata, C.I., R.P. Konstance, P.W. Smith and V.H. Halsinger, 1998. Physical properties of extruded products as affected by cheese whey. *J. Food Sci.*, 63: 814-818.
- Osagie, A.U., 1998. Antinutritional factors. In A. U. Osagie and O. U. Eka (Eds.) *Nutritional quality of plant foods*. Published by Post Harvest Research Unit, Department of Biochemistry University of Benin, Nigeria, pp: 53-83.
- Onimawo, I.A. and S.Osugo, 2004. Effects of germination on the nutrient content and functional properties of pigeon pea flour. *J. Food Sci. and Technol.*, 41: 170- 174.
- Price, L.M. and L.G. Butler, 1977. Rapid visual estimation and spectrophotometric determination of tannin content of sorghum grain. *J. Agric. and Food Chem.*, 25: 1268- 1273.
- Quinn, M.R., L.R. Beuchat, J. Miller, C.T. Young and R.E. Worthington, 1975. Fungal fermentation of peanut flour: effect on chemical composition and nutritive value. *J. Food Sci.*, 40: 470-474.
- Reddy, N.R., S.K. Sathe and D.K. Salunkhe, 1982. Phytates in legumes and cereals. *Advances in Food Research*, 28: 1-92.
- Siegel, A. and B. Fawcett, 1978. Food legume processing and utilization. International Development Research Centre Technical Studies Series, Ottawa.
- Thompson, D.B. and J.W. Erdman, 1982. Phytic acid determination in soybeans. *J. Food Sci.*, 47: 513-517.

- Tomkins, A., D. Alnwick and P. Haggerty, 1987. Fermented foods for improving young child feeding in eastern and southern Africa. In: D. Alnwick, S. Moses and O. G. Schmidt (Eds.), Improving young child feeding in Eastern and Southern Africa. Household Level Food Technology. Ottawa: International Development Research Centre, pp: 198-208.
- Wang, H.L. and C.W. Hesseltine, 1981. Use of microbial cultures: Legume and cereal products. Food Technol., 79-83.
- WHO, 1998. Complementary feeding of young children in developing countries. A review of current scientific knowledge world Health Organisation, Geneva, pp: 130 -131.