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Full Length Research Paper

Evaluation of Quality Characteristics of Composite Diets Prepared From Sprouted and Fermented Millet and Breadfruit Seed Flours

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Four composite diets were prepared from sprouted and fermented millet (*Pennisetum typhoideum*) and breadnut (*Artocarpus communis*) seed flours. The quality was evaluated by determining the proximate composition, microbiological and sensory characteristics. Crayfish and skim milk were added to improve protein quality. The protein and fat contents of the formulated flour samples decreased (10.83% and 4.31%) as a result of sprouting while the crude fibre increased (2.84%). On the other hand, fermentation increased (13.35%, 6.13% and 3.27%) protein, fat and ash contents of the composite diets. During the shelf-life study (21 days), the commercial product (Nutrend) showed minimal growth (from no growth to 1.2×10^3 cfu/g) of microorganism while the composite diets (from no growth to 4.00×10^4 cfu/g). Coliforms and *Escherichia coli* were not detected in any of the samples and the commercial product. Yeast and mould counts were 1.0×10^3 cfu/g to 8.00×10^3 cfu/g and commercial product (2.0×10^2 cfu/g). Sensory evaluation indicated that there were significant differences ($P < 0.05$) between the composite diets and the commercial product in colour, flavor, taste and general acceptability but not in colour ($P > 0.05$).

Keywords: Quality characteristics, composite diets sprouting, fermentation, millet, breadfruit

INTRODUCTION

Complementary food means any food, whether manufactured or locally prepared, suitable as a complement to breast milk or to infant formula. The composition will be close as possible to breast milk except for calories and proteins values (UNICEF, 1998). Complementary foods can be categorized as home foods, fortified foods, or industrially manufactured foods. However, home foods which are less expensive, readily available and culturally acceptable often do not have the nutrient density needed by infant and young children to

satisfy their requirement (Uavy, 2003). Fortification of staple foods, while benefitting the population at large, may not benefit infants and young children because they do not eat enough staple foods for fortification to have an impact. Industrially manufactured complementary foods, although have the potential to improve nutrient intake, their unfamiliarity, lack of availability and possible high cost could discourage the use as complementary foods (Dutta *et al.*, 2006).

Malnutrition is one of the major problems confronting infants and young children in the developing countries. Brock (1961) stated that for every case of kwashiorkor; there were 100 cases of protein malnutrition in the pre-kwashiorkor condition. Several researchers have shown

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that Nigerian children are weaned mainly between the ages of 3-7 months (Ketiku and Smith, 1984; Achinewhu, 1987; Adeyemi *et al.*, 1989). With the widespread problem of infant malnutrition, a lot of interest by both developing countries and international organizations is geared towards the complementation of legume proteins (rich in lysine and tryptophan) with cereal proteins (rich in methionine and cysteine) or supplementation of cereal proteins with legume proteins (Potter and Hotchkiss, 1995; Nwanekezi *et al.*, 2001) so that foods can be produced by mothers, community workers or government agencies and supplied at affordable prices. It is envisaged that this move will go a long way in ameliorating this endemic problem of malnutrition (PAG, 1971). Legumes and cereals which are seeds of domesticated grasses can be sprouted and fermented and used individually or combined to improve the nutritional quality of complementary foods.

The aim of this work was to develop and formulate composite diet blends using sprouted and fermented millet and breadfruit.

MATERIALS AND METHODS

Millet (*Pennisetum tryphoides*) commonly known as "gero", granulated sugar, crayfish, skim milk powder and nutrient (a commercial brand) were purchased at Eke-Ukwu market, Owerri in Imo State. Breadfruit (*Artocarpus communis*), known as "Ukwa bekee" was obtained at Okwelle market in Onuimo Local Government Area, Imo State.

Preparations of sprouted millet and breadfruit seed flours

Millet and breadfruit seed samples each of 200g was cleaned and sorted manually to remove dirt and stones. They were respectively steeped (1:3 w/v) in distilled water in covered plastic containers for 24h (millet) and 120h (breadfruit). Steeping was done for the seeds to imbibe water for easier germination. The steeping water was changed every 12h (millet) and 8hrs (breadfruit). Soaked millet and breadfruit were each sprouted by spreading the seeds on covered moist jute bags for 48h (millet) and 192h (breadfruit). Water was sprayed on each at 12h intervals to keep the germinating seeds moist. The samples were also turned at intervals to ensure proper aeration and the minimization of mould growth. Sprouted millet and breadfruit samples were derooted, dehusked, washed, sorted and dried at 105°C for 1h in a Gallenkamp oven drier. The dried sprouted millet and breadfruit samples were separately milled using disc attrition mill and sifted with a 1mm sieve size to obtain flours of uniform particle size, which were respectively packaged in labeled air tight polyethylene bags and stored at room temperature.

Preparation of fermented millet and breadfruit seed flours

Millet and breadfruit seed samples, each of 200g was manually cleaned and sorted respectively to remove dirt and stones. They were differently soaked in distilled water in covered plastic containers for 24h (millet) and 120h (breadfruit) to allow for anaerobic fermentation. At every 12h, the water used (fermenting water) was differently drained off and replaced. With the fermentation over, the millet and breadfruit seed samples were drained of water, washed, dehusked and dried in a Gallenkamp oven drier at 105°C for 1h. The dried fermented millet and breadfruit samples were separately milled using disc attrition mill and sifted with 1mm sieve size to obtain flours of uniform particle size. The flours were respectively packaged in labeled air-tight polyethylene bags and stored at room temperature.

Preparation of crayfish flour

The crayfish was cleaned, sorted, winnowed and drymilled in an electric blender (National, Model MK 308, Japan). The milled crayfish sample was sifted with 1mm sieve size to obtain the flour. The crayfish flour was packaged in labeled air-tight polyethylene bag and stored at room temperature.

Crayfish contains high protein quality, minerals and vitamins (Mayhew and Penny, 1988).

Preparation of composite diets

Four composite diets were prepared from the sprouted and fermented millet and breadfruit flours. The flour samples consist of varying proportions of millet and breadfruit with sugar, crayfish and skim milk powder in constant proportions (Table 1).

Proximate Composition Analysis

The standard AOAC (2005) method was used to determine the percentage moisture (oven drying at 105°C for 9h), ash, crude fat (solvent extraction) and crude fibre while the Atwater difference method was used for the calculation of total carbohydrate content (Davidson *et al.*, 1975). The protein was determined by the Microkjedahl method (Pearson, 1981). Energy content was calculated using the Atwater quantification of the calorie (energy) contents of food. All determinations were conducted in triplicate and the means \pm SD of three values reported.

Microbiological analysis

Aerobic organisms and yeast, coliform and *Escherichia coli* plate counts were carried out on the sample by the method

Table 1. Formulation of composite diets

Samples	Millet (g)	Breadfruit (g)	Crayfish (g)	Skim milk powder (g)	Sugar (g)	Salt (g)
FMSB	64.00	23.00	5.00	5.00	2.00	1.00
SMSB	55.00	32.00	5.00	5.00	2.00	1.00
FMFB	50.00	37.00	5.00	5.00	2.00	1.00
SMFB	45.00	42.00	5.00	5.00	2.00	1.00

FMSB – Fermented millet – sprouted breadfruit

SMSB – Sprouted millet + sprouted breadfruit

FMFB – Fermented millet + fermented breadfruit

SMFB – Sprouted millet + fermented breadfruit

described by Gilliland *et al.* (1976) after the samples were taken at three (3) days intervals for 21 days (self study). Aerobic Plate Count (APC) was adopted in which standard plate count agar (oxid) was used. One gramme (1g) each of the five (5) composite diets was serially diluted to 10^{-6} . One milliliter (1ml) of each dilution was plated in corresponding duplicate marked plates. Incubation was carried out at 37°C for 48h and the counting of colonies done.

Mould and yeast count was carried out using potato dextrose agar (PDA) (DIFCO, Detroit, MI). Twenty-five grammes (25g) of each sample was blended and homogenated by shaking the mixture. One milliliter (1ml) was pipette into 9ml of buffered peptone water (BPW) and aspirated 10 times with pipette. One milliliter (1ml) of each dilution was plated in corresponding duplicate marked plates containing potato Dextrose Agar (PDA) and allowed to solidify. The plates were incubated at 25°C for 5 days. Colonies were enumerated and counts (cfu/g) obtained. Mould isolates were purified on PDA and then subcultured.

Identification was by the method described by Pitt and Hocking (1977) and Nelson *et al.* (1983).

Escherichia coli count was done with Violet Red Blue Agar (VRB). Twenty-five grammes (25g) of each sample were mixed by shaking with 225ml of buffered peptone water (BPW). One milliliter (1ml) of each solution was pipette into 9 ml BPW. One milliliter (1ml) of each homogenate (dilution) was plated in corresponding duplicate marked plates containing Violet Red Blue (VRB) agar and allowed to solidify. Incubation was carried out at 44.5°C for 48h and the counts done.

Coliform count was carried out with Lauryl Sulphate Broth. Twenty-five grammes (25g) of each blended sample was mixed with 225 ml BPW and homogenated. One milliliter (1ml) of solution was pipetted into 9ml BPW in a tube. Each of the Lauryl Sulphate Broth (LSB) in tubes was inoculated with 1ml of each dilution using sterile pipettes. Incubation was carried out for each of the LSB tubes at 37°C for 48h. Confirmatory test for the coliform bacteria

was done by transferring a loop from each of the LSB tubes into a separate tube of Brilliant Green Bile Agar (BGBA) broth. The tubes were incubated at 37°C for 48h. Gas formation confirmed the presence of coliform bacteria.

Sensory evaluation

Sensory attributes of the four composite diets and a commercial product (Nutrend) were assessed by a 10 man panelist made up of six nursing mothers and four students of the Department of Food Science and Technology, Imo State University, Owerri. The samples were presented without bias in coded white plates and the panelist provided with cups of water at room temperature for mouth rinsing between testing. The following attributes – colour, taste, texture, flavor and general acceptability were rated and scored by the panelists using the 9-point hedonic scale with 9 indicating “Like extremely” and 1 indicating “Dislike extremely” (Ihekoronye and Ngoddy, 1985).

Statistical Analysis

Data on the composite diets were subjected to one-way analysis of variance (ANOVA) and Duncan’s multiple range test was used to separate the means (Steele and Torrie, 1981).

RESULTS AND DISCUSSION

Proximate composition

The result of the analysis on the proximate composition of composite diets produced from millet and breadfruit or breadnut (*Artocarpus communis*) (Table 2) revealed that the ranges of the nutrients of fermented millet – sprouted breadfruit (SMSB), fermented millet – fermented breadfruit (FMFB) and sprouted millet – fermented breadfruit (SMFB)

Table 2. PROXIMATE COMPOSITION OF COMPOSITE DIETS, COMMERCIAL PRODUCT AND THEIR ENERGY (Kcal/g)

Ingredients	Protein (%)	Fat (%)	Ash (%)	Moisture (%)	Fibre (%)	Carbohydrate (%)	Energy (%)
FMSB	11.79±0.01 ^c	7.82±1.08 ^b	3.59±0.78 ^a	8.70±0.70 ^a	1.86±1.11 ^c	66.24±0.62 ^a	382.50 ^b
SMSB	10.83±0.28 ^c	4.31±1.08 ^c	3.03±0.80 ^b	9.30±1.55 ^a	2.84±0.32 ^b	70.05±0.86 ^a	362.31 ^c
FMFB	13.38±1.14 ^b	6.13±0.80 ^b	3.27±0.84 ^b	8.70±0.70 ^a	2.33±0.03 ^b	66.19±0.64 ^a	372.45 ^b
SMFB	14.68±0.91 ^b	5.33±0.77 ^c	4.65±0.67 ^a	9.10±0.87 ^a	2.39±0.66 ^b	63.85±1.55 ^b	362.09 ^c
CP	16.00±0.82 ^a	9.00±0.82 ^a	2.30±0.74 ^c	4.00±1.63 ^b	5.00±0.82 ^a	63.70±1.39 ^b	413.00 ^a

Mean ± SD not followed by the same superscript letter in a column are significantly different ($P < 0.05$)

were protein (10.83 – 14.68%), fat (4.31 – 7.82%), ash (3.03 – 4.65%), moisture (8.70 – 9.30%), crude fibre (1.86 – 2.84%), carbohydrate (63.85 – 70.05%) and energy (362.09 – 383.50kcal) respectively. Sprouted millet – fermented breadfruit (SMFB) diets had the highest (14.68% and 4.65%) protein and ash contents respectively. The protein and fat contents of sprouted millet – sprouted breadfruit (SMSB) composite diets decreased (10.83% and 4.31%) respectively while the crude fibre content increased (2.84%). In terms of fat contents, CP was better than ($P < 0.05$) the other composite diets. FMSB and FMFB diets showed no significant difference ($P > 0.05$) in fat content. The decrease in protein content of pea seed (Beever and Guernsey, 1966) and legumes (Vanderstoep, 1981) by germination had been reported. Akpapunam and Achinwehu (1985) who worked on cow pea (*Vigna unguiculata*) and Youcef *et al.* (1987) on faba beans showed that germination increased the fibre contents of the seeds. Fermentation increased (13.38% and 6.13%) the protein and fat contents respectively but decreased (2.33%) the crude fibre content of fermented millet – fermented breadfruit (FMFB) diets. However, the protein content of commercial product (CP) was significantly different ($P < 0.05$) from other composite diets. FMFB and SMFB diets compared well ($P > 0.05$) with FMSB and SMSB diets. The ash content was decreased (3.72%) by fermentation. The increase in protein content of the fermented formulated sample agreed with the work of Ezeji and Oji-melukwe (1993) on fermented bambara groundnut, fluted pumpkin and millet seed. However, the protein content (16.50%) of the commercial product (Nutrend) was higher than the composite diets. The daily safe levels of protein intake for infants aged 3 months to 2 years ranges from 13.00 – 15.50% (FAO/WHO/UNU (1985). The samples FMFB and SMFB which fell within this range can be regarded as a good protein source for infants and children.

The increase in fat contents of fermented samples may be due to increased activity of the lipolytic enzymes in the fermentation medium which hydrolyzed fat to glycerol and fatty acids (Weiz and Melcher, 1985). There have been reports of similar increase in the lipid level of fermented seeds (Frazier and Westerhoff, 1978; Akpapunam and Achinwehu, 1985). According to PAG (1971), the fat levels should be up to 10.00%. This implied that both the formulated samples and the commercial product (Nutrend) did not fall within the recommendation. However, the low fat content of the composite diets had contributed to their low energy values. The ash content was decreased (3.72%) by fermentation. There was no significant difference in ash content between FMSB and SMFB as well as SMSB and FMFB. The CP was inferior to the other diets. The reduction (3.27%) in ash content by fermentation might be due to the Leaching of some of the inorganic matter into the aqueous medium used for the fermentation which was discarded (Obizoba and Anyika, 1994). The ash content of the nutrend was lower than the composite diets. This meant that the commercial product had low mineral content compared to the composite diets. Both the composite diets and the commercial product (CP) did not fall within the recommendations (5.00%) given by PAG (1971).

The moisture contents of the composite diets were higher (8.70%, 9.30%, 8.70% and 9.10% respectively) than the commercial product (4.00%). Higher moisture content reduces the shelf-life and the quality of food products. Moisture content of any food product is an index of water activity (Uavy, 2003).

The carbohydrate content of the composite diets (FMSB, SMSB, FMFB and SMFB) ranged from 63.85 – 70.05% respectively and were higher than the value (63.70%) of the commercial product (Table 2). The carbohydrate contents of the composite diets were reduced by fermentation. There was no significant difference ($P < 0.05$)

Table 3. TOTAL PLATE COUNT (*CFU/G) OF THE COMPOSITE DIETS AND A COMMERCIAL PRODUCT

DAY	FMSB	SMSB	FMFB	SMFB	**CP
1	No growth	No growth	No growth	No growth	No growth
3	3.00×10^2	2.50×10^2	4.60×10^2	1.10×10^2	No growth
6	4.00×10^2	5.50×10^2	4.60×10^2	2.80×10^2	2.00×10^2
9	9.50×10^2	7.50×10^2	8.50×10^2	4.00×10^2	4.00×10^2
12	1.00×10^2	1.80×10^2	9.50×10^3	5.00×10^2	5.00×10^2
15	1.25×10^4	2.10×10^2	1.00×10^4	7.60×10^2	7.00×10^2
18	3.00×10^2	2.50×10^2	1.20×10^4	8.00×10^2	1.00×10^3
21	4.00×10^2	2.80×10^4	2.00×10^4	1.10×10^3	1.20×10^3

* Colony forming unit

** Commercial product

Table 4. YEAST AND MOULD COUNT OF THE COMPOSITE DIETS AND A COMMERCIAL PRODUCT

FMSB	SMSB	FMFB	SMFB	CP
1.00×10^3	4.00×10^3	1.00×10^3	8.00×10^3	2.00×10^2

in carbohydrate between FMSB, SMSB and FMFB diets. The diets of SMFB and CP compared very well ($P < 0.05$) in carbohydrate contents. The reduction could be attributed to increased activity of α -amylase (Odunfa, 1983a, b) which hydrolyzed starch to simple sugar. From the results, millet had the highest (83.00%) carbohydrate value, followed by SMSB (70.05%) and the least (63.70%) was the commercial product. Sugars provided a source of energy for the fermenting microorganisms. The decrease in carbohydrate content by fermentation agreed with the reports of Isichei and Achinwehu (1988 and 1990) on African oil bean seed and fluted pumpkin seeds and by Nnam (1995) on cowpea.

The food energy values of the composite diets of FMMB, MMMB, FMFB and MMFB respectively ranged from 362.09 – 383.50kcal (Table 2). These values were lower than the commercial product food energy value (413kcal). All fermented processes are accompanied by the liberation of energy in the form of heat. This energy does not account for all the original energy present in the food. This is because part of the energy is used for multiplication and metabolism of microorganisms, so the fermentation process changes the compound of higher energy to those of lower energy (Odunfa, 1983b). The food energy value resulted from the high carbohydrate content. The food energy value includes the fibre content of the carbohydrate (Agu and Aluyah, 2004).

Microbial examination

The microbial analysis of the composite diets (FMSB, SMSB, FMFB and SMFB) respectively are shown in Table 3. The result during the 21 days shelf-study indicated minimal (3.00×10^2 – 4.00×10^4 cfu/g) growth in the samples and it was slightly higher than the values (2.00×10^2 – 1.2×10^3 cfu/g) for the commercial product (Nutrend). The organism identified was *Bacillus* species and their presence showed that the microorganisms survived the drying temperatures. Further, their presence which only served as an indication of unhygienic standard/condition may possibly reduce the shelf-life and quality of the product they contaminate (Gill *et al.*, 1994). The yeast and mould count ranged from 1.00×10^3 – 8.00×10^3 cfu/g (composite diets) and 2.00×10^2 cfu/g (commercial product) (Table 4). Viable plate count was in the order of 10^2 – 10^4 which made the composite diets satisfactory quality under guidelines for ready-to-eat foods (PHLS, 2000). The examination of coliforms and *Escherichia coli* showed that there were no detectable microorganisms (Table 5). The absence of coliforms and *Escherichia coli* implied no faecal contamination in the composite diets.

Sensory attributes

The sensory qualities indicated that there were variations

Table 5. COLIFORMS AND *Escherichia coli* OF COMPOSITE DIETS AND A COMMERCIAL PRODUCT

FMSB	SMSB	FMFB	SMFB	CP
Not detected	Not detected	Not detected	Not detected	Not detected

Table 6. MEAN SENSORY SCORES OF THE COMPOSITE DIETS AND THE COMMERCIAL PRODUCT

Sample	Colour	Taste	Texture	Flavor	General Acceptability
FMSB	5.10 ^b	6.60 ^b	5.90 ^b	6.20 ^b	6.40 ^b
SMSB	4.80 ^b	6.30 ^b	5.90 ^b	6.20 ^b	6.50 ^b
FMFB	5.40 ^b	7.30 ^{ab}	6.30 ^b	6.80 ^{ab}	6.80 ^{ab}
SMFB	6.60 ^{ab}	8.30 ^a	6.90 ^b	7.40 ^a	8.00 ^a
CP	6.90 ^a	6.10 ^b	8.30 ^a	6.80 ^{ab}	7.30 ^{ab}

Mean ± SD not followed by the same superscript letter in a column are significantly different (<0.05)

in the attributes of the composite diets (Table 6). In terms of colour, sprouted millet – fermented breadfruit (SMFB) and commercial product (CP) did not differ significantly ($P>0.05$) and were mostly preferred. However, the diets of FMSB, SMSB and FMFB compared well ($P>0.05$) in colour. There was significant difference ($P<0.05$) in texture between CP and the composite diets but differed significantly ($P<0.05$) in flavor from FMSB and SMSB diets. Diet SMFB was significantly different ($P<0.05$) from other composite diets and CP in taste. Furthermore, FMSB and SMSB diets compared well ($P>0.05$) in flavor and general acceptability.

CONCLUSION

The study showed that composite diets can be formulated from blends of sprouted and fermented millet and breadfruit (*Artocarpus communis*) seed flours. Sprouting and fermentation enhanced the protein, fat, crude fibre and ash contents of the diets. Furthermore, Coliforms and *Escherichia coli* were not detected, indicating the absence of contamination of the composite samples.

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