EVALUATION OF NUTRITIONAL QUALITY OF AN INFANT WEANING FORMULA FROM UNDERUTILIZED PROTEIN SOURCE

*[1]PETROL B. B., [2]MOHAMMED Z. K., [1]UMAR M. U. AND [3]HASHIM H.

[1]Department of Biochemistry, University of Maiduguri, Nigeria
[2]Department of Biochemistry, Kaduna State University
[3]Department of Biochemistry, Yobe State University.

Corresponding Author: bintubukar@yahoo.com

ABSTRACT

Cereal/legume/tuber vegetable infant weaning diet was produced using in a 60:20:20 ratio. The yellow maize was fermented for 72hours to produce Akamu. Standard methods were used for the determination of pH, titratable acidity, chemical composition and *in vitro* protein digestibility of the infant weaning diet. Fermentation was found to cause a reduction in pH with time, in a pH drop from (5.76 to 3.5). The infant weaning diet had a moisture content of (2.01±0.07%) and (frisocream®) had (2.0%). Result of the crude protein of the infant weaning diet (16.9±0.03%) was slightly higher than Frisocream® (15.3%). The carbohydrate of the infant weaning diet (82.3± 0.06%) was below the values of Frisocream® (445%). The *in vitro* protein digestibility of the infant weaning diet significantly increased with time. Therefore, the infant weaning diet had a moisture and protein content comparable to frisocream.

Keywords: Infant weaning Formula, Tiger nut flour, Cowpea flour, Chemical Composition.

INTRODUCTION

Weaning has been described as the substitution of the mother's milk with other foods. It is a gradual process by which an infant is introduced to adult diet. Weaning food is therefore a special formulation, which is a supplement to the breast milk. The report by (Wakil and Kazeem, 2012) indicated that breast milk, even from well-nourished mothers, might be inadequate to meet the nutritional needs of the infant after the first three months of life, hence the need for a supplementary or weaning food. This weaning period is a very critical period in the life of a child and if not well managed might lead to malnutrition and other health implications (Anigo *et al.,* 2010). In many developing countries, children, especially those in the low-income class, are weaned on cheap, readily available starchy foods that are very poor in protein and other essential nutrients (Onilude *et al., 2014*).

In various parts of Nigeria, children weaned between the age of 3-24 months, are mostly fed on local staples like pap from corn sorghum, that do not meet the nutritional requirements of the infant (Ijarotimi, 2012). Many studies have reported on supplementation of cereals and other carbohydrate-based meals, however the problem of malnutrition still persists in many communities of sub-saharan African. This is attributable to insecurity and its impact on food security. Several factors that include poor nutritional education, decline in income and high cost of commercial weaning foods add to the burden of malnutrition in developing countries. (Nkama *et al.,* 2001).

This study was designed based on the background that to prepare a weaning food formulation using locally available and cheap raw material like cowpea, maize (yellow) and tiger nut. The nutritional properties of the formulation were evaluated and compared with commercially available weaning food as well as its ability to meet recommended dietary allowance (RDA). For children and could serve as alternative or supplementary sources during the critical phase of weaning.

MATERIALS AND METHODS

Sources of Materials

The yellow maize, cowpea and tiger nut were purchased in Maiduguri, Monday Market and authenticated by a plant taxonomist in the department of Biological sciences University of Maiduguri.

Preparations of Akamu

The ‘akamu’ (Ogi) was prepared by the method described by Akingbola *et al* (1981). One hundred grams (100g) of the cleaned pearl millet (raw grain) was cleaned and steeped in 200cm^3 of distilled water (1:2 ratios) for 72 hours. At the end of the 72 hours, the tap water decanted and 200cm^3 of distilled water was added and milled with warring blender for 4min at rheostat setting of 120. The slurry obtained was sieved through a nylon cloth to separate the bran after adding 6000 cm^3 of distilled water. The ‘kamu’ was then allowed to stand for 24 hours for the starchy part to settle, after which the water was decanted and the kamu was sun dried to a constant weight.

Preparation of Cowpea

One hundred grams (100g) of the cowpea was cleaned of dirt and soaked in distilled water for 20 minutes. The cowpea was then dehulled using a pestle and mortar. It was then washed to separate the husk, after which it was dried to a constant weight. The cowpea was ground to fine powder (Theodore *et al.,* 2007)

Treatment of Tiger Nut

The tiger nut was washed with distilled water to remove dirt. The washed tubers when the soaked in 1% (W/V) sodium carbonate and left to say overnight at pH 10.0 the tiger nut were than washed and resoaked in 1% (W/V) citric acid at pH of 3.0 and left to overnight, after which acid was drained (Rosello-Soto *et al.,* 2019)

Formulation of the Infant Weaning Formula

Cereal/legume diet was formulated using in a 60:20:20 ratio i.e Yellow Maize 60g, Tiger nut 20g, Cowpea 20g. Cowpea seed that are treated were mixed together and pulverized in a mortar and then homogenized in a blender. The mixture was then strained through two layers of white cotton cloth and the emulsion were squeezed out, the meal was then blanched and poured on an aluminum tray covered with aluminum foil. The milk was evenly distributed on the tray and placed in an oven to dry for 48 hours at a temperature of 70^{o}C. The dried milk which was light brown in colour was ground in to a powder form.

pH and Titratable Acidity

The pH of the samples was determined according to the method of AOAC (2010) the titratable acidity was estimated by titrating against 0.1 N Na0H to phenolphthalein end point and the acidity was calculated as lactic acid/100g.

Proximate Composition

The samples were analyzed for moisture content, dry matter, crude protein, crude fiber, either extract or fat, ash, carbohydrate and nitrogen free extract, (NFE) according to AOAC method 2010 15th edition.

Estimation of Moisture Content

The sample was dried to constant weight at 70^{o}C in an oven. The difference in the weight of the sample is the moisture content of the sample.

Estimation of Dry matter

The dry matter content of the sample was determined by weighing 10g of samples into the Petri dish while placed in hot oven at temperature of 105^{o}C for 24 hours. And then removed and placed in desiccator to cool, after cooling it was weighed. The dry matter content was calculated using formula.

$MC = (w_1\text{-}w_2/w_2\text{-}w_i) \text{ X } 100$

Where:

W_1: weight of Petri dish with sample in gram before oven dried
W_2: weight of Petri dish with sample in gram after oven dried.
W_i: weight in gram of empty Petri dish.

Estimation of Crude Protein Content

The crude protein content was analyzed using keljebal tablets and 1g or 2g of samples was weight into a digested tube 1 or 2 Keijebal tablets were added, 10 or 20mLs of concentrated sulphuric acid (conc. H_2SO_4) was added onto the tube and digested at 420°C for 3 to 5 hours. After cooling 80 or 9mLs of distilled water was added onto 50mLs of digested and diluted solution and then placed on heating section of the distillation chamber, 30mLs of 4% boric acid, plus bromocresol green and methyl red as an indicator was put onto conical flask and placed underneath the distillation chamber for collection of ammonia, the solution changed from orange to green colour about 0.1 normal solution of hydrochloric acid (HCL) was weight into burrete. The conical flask containing the solution was titrated until the colour changes from green to pink. The burrete reading was taken the crude protein was calculated using the formula.

$$\%CP = \frac{(A-B)NxFx6.25x100}{Mg\ of\ samples} X\ 100$$

Where:
A: Mis of acid used for titrating the samples
B: Mis of acid used for titrating blank sample (0)
N: Normality of acid used for titration.
F: Factor 14.007
6.25: Is constant
100: Conversion to percentage

Estimation of Crude Fiber

The crude fiber was determined by weighing 2g of samples was placed in a round or flat bottom flask and 50mLs of tri-chloroacetic acid reagent (TCA) was added the mixture was boiled and refluxed for 40 minutes. Filter paper was removed and cooled to temperature. Filter paper was used to filter the residue. The residue obtained was washed 4 times with hot water and once with petroleum either then the filter paper plus the sample were folded together and dried at 30°C-60°C in an oven for 24 hours reweighed and then ashed at 650°C and then coaled reweighed.

$$\%CF = \frac{difference\ in\ weighing}{weigh\ of\ sample\ on\ Dm\ basis} X\ 100$$

Estimation of Ether Extract (Fat) Content

Estimation of the ether extract was determined by using 80xhlet apparatus, 1 or 2g of the feed sample was weight into a thimble and 200mLs of petroleum ether was measured with measuring cylinder. The solution was put into round or flat bottom flask and was heated at 45°C for 1 hour interval for 2 hours. The collecting flask and was removed and cooled into dissecator for 15 minutes and percentage fat sample is determined by using the formula.

$$\%fat = \frac{weight\ of\ fat}{Weight\ of\ sample} X\ 100$$

Estimation of Ash Content

To determine the ash content 1 or 2g of sample was weighed into crucible and tried at 105⁰C for 24hour, then cooled in the desiccators for 15 minutes and reweighed, it was then charred at 600⁰C or 650⁰C in muffle furnace for 2-3 hours then cooled in desiccators for 15 minutes and reweighed Formula:

$$\%ASH = \frac{loss\ in\ weight}{initial\ weight} X\ 100$$

Estimation of Nitrogen Extract

Percentage nitrogen free extract was determined by computing indirectly by difference using the formula:

%NFE = 100 – (%CP+%CF+%EE+%ASH)

Estimation of Carbohydrate

Percentage carbohydrate was determined by computing indirectly by difference using the formula:

%Carbohydrate = 100 – (%MC+%ASH+%CF)

In vitro protein digestibility (IVPD)

The *in vitro* protein digestibility of the sample was determined by enzymatic method (Nills, 1979). A known weight of the sample, containing 16 mg nitrogen was taken in triplicate and digested with 1mg pepsin in 15 ml of 0.1m HCl at 37°C for 2 hours. acid (TCA). The mixture was then filtered quantitatively through what No. 1 filter paper. The TCA soluble fraction was assayed for nitrogen using the micro – Kjadahl method protein digestibility of the sample was calculated by the following formula.

$$\text{Protein digestibility (\%)} = \frac{\text{N in supernatant} - \text{blank N}}{\text{N in sample 8}} \text{X } 100$$

Statistical Analysis

All determination was carried out in triplicates. All data collected was subjected to analysis of variance and Duncan multiple range test was used to compare the means.

RESULTS

Table 1 shows the pH and titratable acidity of yellow maize, tiger nut and cowpea and values were statistically different (P<0.05). A reduction of pH from 5.76 ± 0.15 to 3.5 ± 0.05 was observed for the 72hours fermentation of yellow maize while an increase from 1.06±0.11 to 2.5±0.10 was observed for the titratable acidity at 72hours fermentation.

Table 1: pH and Titratable Acidity of Yellow Maize at 72 hours of Fermentation.

Hours	Sample	pH	TA(glactic acid/100g)
0	Yellow maize	5.76 ± 0.15^{a}	1.06 ± 0.11
24		5.0 ± 0.11^{b}	0.96 ± 0.11^{a}
48		4.1 ± 0.15^{c}	1.9 ± 0.17^{b}
72		3.5 ± 0.05^{d}	2.5 ± 0.10^{c}

Values are mean ± standard deviation of three determining 60g yellow maize, 20g tiger nut and 20g cowpea. Means with different superscripts in a column for a sample are significantly different (p<0.05).

Table 2 shows the Proximate Composition of unprocessed and Processed Yellow maize, Tiger nut , Cowpea. There was a significant reduction in the moisture content of processed tiger nut (3.45±0.06), yellow maize (3.70±0.20) and cowpea (3.70±0.04) as compared to the unprocessed tiger nut (4.00±0.02), yellow maize (4.59±0.04) and cowpea (4.27±0.06). Crude protein content of processed tiger nut had a value of (2.97±0.01), yellow maize (4.70±0.02) and cowpea (5.56±0.09) while the unprocessed samples had a protein content of (2.48±0.03), (3.18±0.03) and (5.14±0.10) respectively. No significant increase was observed in the carbohydrate content of unprocessed and processed yellow maize and cowpea, however there was a significant increase in the carbohydrate value of processed tiger nut (86.23±0.30) as compared to unprocessed tiger nut (70.28±0.36). Processed yellow maize and cowpea showed higher values in the fiber content than the unprocessed samples. The fat content of the infant weaning formula (9.94±0.06) was close to that of the commercial weaning food frisocream® (13.2).

Table 3, proximate composition of the infant weaning diet blend compared with a commercial weaning meal frisocrem® (rice) mg/100g. The moisture content of the infant weaning formula (2.01 ±0.07) had the same value with the commercial weaning food frisocream® (2.0). The value of protein of the infant weaning formula (16.9±0.03) met that of the commercial weaning food frisocream® (15.3).

Table 2: Proximate Composition of Yellow Maize Tiger nut and Cowpea.

Proximate analysis (%)	Tiger nut		Yellow Maize		Cowpea	
	Unprocessed	Processed	Unprocessed	Processed	Unprocessed	Processed
Dry Matter	95.99 ± 0.01	96.49 ± 0.07	95.20 ± 0.34	95.05 ± 0.012	95.61 ± 0.10	95.73 ± 0.67
Moisture	4.00 ± 0.02[a]	3.45 ± 0.06[a]	4.59 ±0.04[c]	4.70 ± 0.20	4.27 ± 0.06[a]	3.70 ± 0.04[f]
Protein	2.48 ± 0.03[a]	2.97 ± 0.01[a]	3.18 ± 0.03[c]	4.70 ± 0.02[a]	5.14 ± 0.10[e]	5.56 ± 0.09[f]
Fat	2.99 ± 0.01[a]	3.10 ± 0.14	3.99 ± 0.01[b]	4.03 ± 0.06	2.21 ± 0.18[a]	3.06 ± 0.12[b]
Crude Fiber	18.84 ± 0.21[a]	20.24 ± 0.22[b]	3.98 ± 0.02[c]	2.12 ± 0.11[a]	1.98 ± 0.02[e]	2.10 ± 0.09[d]
Ash Content	4.06 ± 0.12[a]	4.82 ± 0.23[a]	0.98 ± 0.01[b]	0.99 ± 0.01[d]	0.98 ± 0.02[b]	0.98 ± 0.02[b]
Carbohydrate	70.28 ± 0.36[a]	86.23 ± 0.30[a]	87.39 ± 0.21[b]	87.34 ± 0.05[b]	87.6 ± 0.06[b]	87.71 ± 0.02[b]

Values are Means ± SD of triplicates
Values with different super script in a column are significantly different P<0.05

Table 3: Proximate Composition of the Weaning Diet Compared with a Commercial Weaning Food Frisocrem® (rice)

Proximate analysis %	Yellow maize, Tiger nut and Cowpea (60:20:20 mg/100g)	Commercial Weaning Food Frisocream®
Dry matter	94.65 ± 0.13	ND
Moisture	2.01± 0.07	2.0
Crude protein	16.9 ± 0.03	16.3
Ether Extract	9.94 ± 0.06	13.2
Crude fiber	4.94 ± 0.06	ND
Ash	2.98 ± 0.02	ND
Carbohydrate	82.3 ± 0.06	445

Value are recorded as mean ±SD of three determining.
ND-Not Determined

Table 3. Shows the *in vitro* protein digestibility of unprocessed and Processed Yellow Maize, Tiger nut and Cowpea and the values were statistically different (P<0.05).

The *in vitro* protein digestibility of processed, yellow maize, tiger nut, cowpea and the infant weaning diet significantly increases with time from 1 hour to 6 hours.

Table 4: *in vitro* protein digestibility of Raw and Processed Yellow Maize, Tiger nut and Cowpea.

Digestibility	Tiger nut (%)		Yellow Maize (%)		Cowpea (%)		IWD
Time	unprocessed	Processed	unprocessed	Processed	unprocessed	Processed	60:20:20
1 hour	40.4±0.30[a]	49.3±0.20[a]	25.9 ±0.23[a]	36.9 ± 0.29[a]	45.78±0.26[a]	50.4±0.29[a]	60.28±0.06[g]
6 hours	56.4±1.9[c]	65.1±3.5[c]	48.5 ± 0.40[c]	53.4 ± 0.39[c]	70.3±0.40[c]	76.0±0.50[c]	87.33±0.61[h]

Values are mean ± standard deviation of triplicates (60g yellow maize, 20g tiger nut and 20g Cowpea). Means with different superscripts in a column are significantly different (p<0.05).
IWD- Infant Weaning Diet.

DISCUSSION

Tiger nut and Cowpea has been found to cause a reduction in pH with time. Similar results were also found by Singh *et al*., (2012). These results also agree with those obtained by Giese (1994) who reported that as a result of fermentation acidity increase and pH falls down and this enhance the Keeping Quality of the foods by inhibiting microbial growth Usha and Chandra (1997). This result is in agreement with Wakil and Kazeem, 2012) who reported that lactic acid fermentation causes a rapid drop in pH of various food grains.

The proximate composition of the infant weaning diet had low moisture content as that of the commercial weaning food. High moisture content of food encourages microbial growth. (Oyeruka, 2011). This is an important consideration in local feeding methods in Nigeria, because most mothers often prepare large quantities of dry infant foods and keep in containers, to avoid frequent processing, in order to have spared time and energy for other domestic activities. The slight decrease in fat as compared with to commercial weaning meal of frisocrem® (rice) tallies with earlier report of Laminu *et al*., 2014. The protein content of the infant weaning diet compared with the commercial weaning food frisocrem® (rice) could be due to the method of processing of weaning diet and incooperation of tigernut and cowpea. Ikejenlola and Adurtoye, 2014 reported that high protein in infant diet may be due to the use of legume as fortifying agent and has been reported to improve the protein content of cereal-based diets.

The carbohydrate content of the infant weaning diet also shows a significant decrease as compare to commercial food but tend to be very close; this also may be due to the method of processing weaning diet. The increase in *in vitro* protein digestibility of produced infant weaning diet was as a result of processing Microflora may produce proteolytic enzymes during fermentation which may be responsible for the increase of protein digestibility. In addition, the elimination of phytic acid contributes to the improvement in protein digestibility. These results agree with (Mohiedeen *et al.,* 2010) who reported that fermentation was found to improve the *in vitro* protein digestibility and this could be attribute to the partial degradation of complex storage proteins into simpler and more soluble product.

CONCLUSION

The results indicate that the infant weaning diet produced from yellow maize, tiger nut and cowpea had a low moisture and high protein content which was comparable to frisocream indicating that the underutilized crops can be used to produce infant weaning diets. It is recommended that further studies on functional properties and microbial analysis should be carried out.

REFERENCES

Akingbola, J.O, Roorey, L.W. and Fallbion, J.M. (1981). A laboratory Procedure for the Preparation of Ogi, a Nigerian Fermented Food. *Journal of food siencec,* 64: 1523-1526.

Anigo, K.M., Ameh, D.A., Ibrahim, S. and Danbauchi, S.S. (2010). Nutrient Composition of Complementary Food Gruel Formulated from Malted Cereals, Soy Beans and Groundnut for use in North-Western Nigeria. *African Journal of Food Scienc,e* 4 (3): 65-75.

AOAC (2010). Association of Official and Analytical Chemist. Official Methods of Analysis. Washington DC.Vol II. No. 44, 16th Edition pp 22-33.

Davies N.T. and Raid, H. (1979). An Evaluation of phytate zinc availability form soya based textured – vegetable protein meat substitutes or meat extenders. *British Journal of Nutrition.* 41:579.

Ijarotimi, O.S., Keshino O.O. (2013). Determination of Amino acid, Fatty Acid, Mineral Functional and Choking Properties of Germinated and Fermented Popcorn (zea mays) flour. *In European Journal and of Food Research,* and review vol. 2011, No. 2. P102 – 122.

Ikujenlola, A.V, Adurtoyo E.A. (2014). Evaluation of Quality Characteristics of high Nutrient Dense Complementary of from Mixtures of Malted Quality Protein Mazie and Steamed Cowpea. *Journal of food process technol,* 5:29.

Laminu, H.H. Sheriff M. Bintu B.P. and Muhammad A.A. (2014). Evalution of the Protein Quality of Composite Meals Produced from Selected Cereals and Legumes of Infants. *Scholarly journal of agriculture science,* Vol. 4(11) pp. 536 – 542.

Mohiedeen, I.E., Alt,.E., Tinay, A.E,O., Elkhayaie. E., Babiker, H. and Mallasiy,I.O.(2010). Effect of Fermentation on *in vitro* Protein Digestibility Protein Fractions and Acids Composition of Maize (zeamays) *cultivar). Journal of Enviromental Agriculture. Food Chemistry,* 9 838-847.

Nkama I, Dagawanna F.N. and Ndahi, W.B. (2001). Production Composition and Consumer Acceptability Weaning of Foods from Mixtures of Pearl Millet Cowpea and Groundnut. *Journal of Arid Agric,* 11:165 – 169.

Onilude A.A., Sanni A.I. and Igholo M.I. (2014). Process Upgrade and the Microbiological, Nutritional and Consumer Acceptability in Infant Weaning Food from Fermented Composite Bends of Cereals and Soybean. *Journal of food, Agriculture and Environmental Science*, 2:64 – 68.

Oyarekua, M.A. (2011). Sensory Evaluation, Nutritional Quality and Antinutritional Factors of Traditionally Co-Fermented Cereals / Cowpea Mixture as Infant Complementary of Food. *Agric Biology Journal of North America,* IC5: 950 – 956.

Singh, A, Yadav, N. and Sharma, Shouti. (2012). Effect of Fermentation on Physicochemical Properties and *in vitro* Starch and Protein Digestibility of Selected Cereals. *International journal of agricultural and of food science,* vol. 2(3) pp. 66 – 70.

Theodore, I., Mbata, M.J. Ikeheboneh, I. and Ahonkai, S., (2007). Nutritional Status of Maize Fermented Meal by Fortification with Bambara nut. *African Journal of Food and Nutrition Development,* 7(2). 44-61.

Usha A and Chandra, T.S. (1997). Microbial Population and Biochemcail Changes in Fermenting Finger Millet *(Eleusine coraeana). World journal of microbiology and Biotechnology*, 13: 533 – 537.

Wakil S.M. and Kazeem, M.O. (2012). Quality Assessment of Weaning of Food Produced from Fermented Cereal Legume Blend using Ttarter. *International of food research journal,* 19(4): 1679 – 1685.