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Comparative Evaluation of the Nutritional Quality, Functional Properties and Amino Acid Profile of Co-Fermented Maize/Cowpea and Sorghum/Cowpea *Ogi* as Infant Complementary Food

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Abstract: This study involved formulating nutritionally suitable complementary food mixtures with locally available raw materials. Maize or sorghum was mixed with cowpea, soaked at 25°C for 72 h, wet-milled and sieved. The sediment was sun dried, milled for analyses. Proximate, functional properties and amino acid were determined in co-fermented maize/cowpea and sorghum/cowpea. Sorghum/cowpea had higher water absorption capacity, (235%) than maize/cowpea (103%) sorghum/cowpea and a lower value of oil absorption capacity (47.9%) than, maize/cowpea of (67.6%). Oil absorption capacity of (14.7%) in sorghum/cowpea was higher than (9.6%) in maize/cowpea. The higher foaming capacity of maize/cowpea (40.0%) than that of sorghum/cowpea of 20.0% might be due to soluble proteins and higher emulsion capacity of maize/cowpea might make it a better flavour retainer and enhanced mouth-feel. Values of foaming stability, least gelation capacity and bulk density (loose and packed) were comparable. Sorghum/cowpea had higher contents of lysine, histidine, arginine, aspartic acid, threonine, serine, alanine, glutamic acid, proline, glycine, cystine, valine, isoleucine, leucine, tyrosine, phenylalanine, total amino acids, total sulphur amino acid, ratio of total essential amino acids/aromatic amino acids but lower values of methionine and total essential amino acids. Thus co-fermented sorghum/cowpea is of better protein quality than maize/cowpea.

Key words: Co-fermentation, functional properties, proximate composition, amino acid

INTRODUCTION

Cereals and cowpea are widely used food crops in the developing countries and their importance is increasing as complementary foods for infants. Fermented cereal has been popularly used as complementary infant food in Nigeria, where it is referred to as *ogi*. *Ogi* is a smooth, creamy, free-flowing thin porridge obtained from wet-milled, fermented maize, sorghum or millet. The preparation involves dilution of the fermented product with water and boiling with constant stirring to desired consistency (Johansson *et al*., 1995). Complementary fermented gruel is consumed by over 90% of children over 6 months of age in Nigeria. Starch and protein are reported to be the major components affecting function properties of food material. Fleming *et al*. (1974) reported that water absorption capacity is attributed to protein content of food material. According to Sefa-Dedeh and Afaokwa (2001), addition of cowpea improved the water absorption potential of fermented maize dough and that protein is responsible for the bulk of water uptake and to a lesser extent the starch and cellulose at room temperature.

According to Chauvan and Kadan (1989), processes like fermentation lead to modification in functional properties and has been used to enhance desirable properties for food formulations. Fermentation tends to influence the functional properties of foods. Protein and carbohydrates undergo

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significant hydrolytic changes during fermentation. Natural fermentation tend to influence the functional properties and might lead to desirable or undesirable modification of functional properties. Ahmed *et al*. (1988) reported that fermentation increase protein which is reflected in better water absorption capacity, water retention capacity and fat absorption capacity. Fermentation is also reported to improve foaming properties (higher foaming capacity corresponds to poor foaming characteristics) but not emulsion stability.

Bulk density of meal is also influenced by number and packing density of protein bodies and starch granules. High protein content and protein-lipid interactions were said to contribute to increase in fat absorption capacity of food. Starch and soluble proteins and other seed components influence foaming capacity and stability and emulsifying properties. Various studies have been done on supplementation of Nigerian *ogi* with cowpea. Akpapunam and Sefa-Dedeh (1995), Sefa-Dedeh and Afaokwa (2001) and Soulski and Summer (1987) reported that utilization of cowpea products depends on its functional properties and that cowpea products are superior to soybean in terms of water absorption capacity especially the protein isolates however, there is scanty information on co-fermentation of maize, sorghum and cowpea as complementary infant food. This study is reporting the comparative effect of co-fermentation on the functional properties of co-fermented maize/cowpea and millet/cowpea mixtures and their amino acid profiles.

MATERIALS AND METHODS

Materials

Sample of fermented sorghum/cowpea *ogi* and co-fermented maize/cowpea *ogi* flours.

Proximate Composition

This was determined according to AOAC (1980).

Reducing Sugars

About 1.3 mL of fermented or non-fermented slurry was centrifuged at 5000 g min^{-1} for 10 min, 0.2 mL 15 N NaOH was added to the supernatant in each tube, vortexed and stored in the freezer at -20°C for further use.

The tubes were later thawed and 1 mL of each tube was diluted with 1 mL Dinitro-Salicylic Solution (DNS). Each mixture was agitated for 5 minutes in water bath at 100°C. The tubes were then rapidly cooled in ice. They were then rapidly cooled in ice. They were later vortexed and read at 575 nm.

Bulk Density

Bolk density of loose and packed flour were determined according to the method of Wang and Kinsella (1976).

Water Absorption Capacity (WAC)

This was determined according to the method of Soluski (1962).

Fat Absorption Capacity

This was determined according to the method of Soulski (1962).

Foaming Capacity

Was determined according to the method of Sathe *et al*. (1982).

Emulsification Capacity

This was determined according to the method of Beuchat *et al*. (1975).

Least Gelation Capacity

This was determined according to the method of Coffman and Garcia (1977).

Determination of Amino Acids Profile

The amino acid profile in the samples was determined using methods described by Speckman *et al*. (1958). The samples was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and located into the Technicon Sequential Amino Acid Analyzer (TSM).

Defatting of Samples

A known weight of the samples were dried into extraction thimble and the fat was extracted with chloroform/ methanol (2:1 mixture) using Soxhlet extraction apparatus as described by AOAC (1990). The extraction lasted for about 15 h.

Hydrolysis of Samples

A known weight of the defatted sample was weighed into glass ampoule. A 7 mL of 6 N HCl was added and oxygen was expelled. The glass ampoule was then sealed with Bunsen burner flame and put in an oven preset at 105±5°C for 22 h. The ampoule was allowed to cool before broken opened at the tip and the content was filtered. The filtrate was then evaporated to dryness at 40°C under vacuum in a rotary evaporator. The residue was dissolved with 5 mL of acetate buffer (pH 2.0) and stored in plastic specimen bottles which were kept in the freezer.

Loading of the Hydrolysate into TSM Analyzer

About 5 to 10 µL was dispensed into the cartridge of the analyzer for 76 min. Amino acid values was calculated using the chromatogram peaks; while the net weight of each peak produced by the chart record TSM was measured while the half-height of the peak was accurately measured and recorded. Approximate area of each peak was then obtained by multiplying the height with the width at half-height.

The Norleucine Equivalent (NE) for each amino acid in the standard mixture was calculated using the formula:

$$NE = \frac{\text{Area of Norleucine peak}}{\text{Area of each amino acid}}$$

Protein Efficiency-Ratio (PER) is calculated as: PER= -0.468+0.454(leucine)-0.105 (tyrosine).

RESULTS

In Table 1, the dry matter contents were higher in unfermented samples than in co-fermented samples. The pH of the fermenting medium decreased over 72 h; however the decrease was more significant in sorghum/cowpea. The crude protein values were comparable in both co-fermented

Table 1: The chemical proximate composition of co-fermented sorghum/cowpea, co-fermented millet/cowpea *ogi* and unfermented samples

Sample	Dry matter	pH	TTA	Crude protein	Crude fibre	Lipid content	Reducing sugars	Ash
LMZ/C	86.9±0.05^{ab}	5.4±0.05^{e}	0.27±0.00^{cde}	13.9±0.04^{e}	4.3±0.10^{abc}	1.3±0.06^{a}	0.28±0.02^{ab}	2.30±0.05^{c}
UMZ/C	89.3±0.04^{de}	6.4±0.01^{f}	0.10±0.00^{abc}	13.4±0.35	5.6±0.4^{bc}	4.8±0.21^{cd}	0.12±0.30^{a}	3.60±0.06^{g}
LS/C	86.1±0.00^{ab}	4.5±0.00^{c}	0.14±0.00^{abcd}	13.4±0.00	4.0±0.00^{abc}	2.9±0.04^{b}	0.60±0.03^{abc}	0.60±0.00^{a}
US/C	88.1±0.015^{ab}	6.5±0.03^{i}	0.08±0.01^{a}	13.8±0.10	4.9±0.33^{abc}	4.1±0.10^{d}	0.21±0.21^{ab}	2.09±0.00^{f}

Each value is mean of triplicate values. values with the same superscript in each column are not significantly different ($p>0.05$). TTA = Total titrable acidity and unfermented samples. The ash, crude fibre and lipid values were higher in unfermented samples than in co-fermented samples but the crude fibre values of the co-fermented samples were comparable. However, the value of co-fermented sorghum/cowpea was significantly ($p<0.05$) higher (2.9 g/100 g), than that of co-fermented maize/cowpea with a value of 1.3 g/100 g. Co-fermented maize/cowpea, 0.23 g/100 g and sorghum/cowpea 0.60 g/100 g.

In Table 2 the water absorption capacity value was higher ($p<0.05$) in sorghum/cowpea *ogi* (235%) than maize/cowpea *ogi* (103.5%). Also, the oil absorption capacity value was lower in sorghum/cowpea with a value of 47.9 *ogi* than maize/cowpea *ogi* with a value of 67.6%; the oil absorption stability was also higher in sorghum/cowpea *ogi* (14.7%) than maize/cowpea with a value of 9.6%.The foaming capacity was higher ($p<0.05$) in sorghum/cowpea *ogi* with a value of 40% than maize/cowpea *ogi* with a value of 20%; while the foaming stability values were comparable in maize/cowpea (1.3%) with sorghum/cowpea o*gi* (1.3%). The value of emulsification capacity was higher ($p<0.05$) in maize/cowpea *ogi* with a value of 15% than sorghum/cowpea *ogi* with a value of 5%.The least gelation capacity values were comparable in both samples with a value of 5.0% each. The values for bulk density (loose flour) were comparable for sorghum/cowpea *ogi* and maize/cowpea *ogi* while the bulk density values for packed flours was higher in sorghum/cowpea than maize/cowpea *ogi*.

Table 3 shows the amino acids composition of co-fermented maize/cowpea and sorghum/cowpea. Sorghum/cowpea had higher values of lysine (24.3), Aspartic acid (68.8) and glutamic acid (200),

Table 2: The values of functional properties of sorghum/cowpea *ogi* and co-fermented maize/cowpea *ogi*

Functional property (%)	Sorghum/cowpea *ogi*	Maize/cowpea *ogi*
WAC	235.00	103.50
OAC	47.90	67.60
OAS	14.70	9.60
FC	20.00	40.00
FS	1.30	1.30
EC	5.00	15.00
LGC	5.00	5.00
Bulk density (loose) cm^3	0.75	0.71
Bulk density (packed flour) cm^3	1.10	0.71

WAC: Water absorption capacity, OAC: Oil absorption capacity, OAS: Oil absorption stability, FC: Foaming capacity, FS: Foaming stability, EC: Emulsion capacity, LGC: Low gelation capacity

Table 3: Amino acid content (mg g^{-1}) of co-fermented Maize/cowpea and Sorghum/cowpea *ogi*

Amino acid	Maize/cowpea	Sorghum/cowpea
Lysine	36.30	42.30
Histidine	20.90	22.80
Arginine	36.30	44.30
Aspartic acid	57.50	68.80
Threonine	29.40	32.90
Serine	40.10	45.20
Alanine	70.80	81.80
Glutamic acid	170.00	200.00
Proline	66.90	88.90
Glycine	25.00	31.80
Cystine	19.90	21.30
Valine	46.00	52.90
Methionine	19.60	20.90
Isoleucine	35.90	38.20
Leucine	101.00	129.30
Tyrosine	30.40	40.30
Phenylalanine	41.90	51.50
TAA	879.00	993.00
TEAA	407.00	342.50
TEAA/TAA	0.46	0.34
TSAA	39.50	42.20
ArAA	72.30	92.00
P-PER (g/100 g)	3.90	7.20

TAA: Total amino acids, TEAA: Total essential amino acids, TSAA: Total sulphur amino acids, ArAA: Aromatic amino acids. P-PER: - 0.468+0.454 (leucine) -0.105 (tyrosine) glycine (31.88), alanine (81.8), leucine (129.3), isoleucine (38.2), cystine (21.3) phenylalanine (51.5), proline (8.89), tyrosine (4.03) histidine (22.8),threonine (32.9), serine (45.2) and valine (52.9) than in maize/cowpea with methionine (19), aspartic acid (5.70), glutamic acid (171.0), glycine (25) alanine 70.9) leucine (101.1) phenyl alanine (55), valine (52.9) and proline (Glutamic acid was the most abundant in sorghum/cowpea and maize/cowpea *ogi* (66.9) followed by proline and aspartic acid. The values of cystine and histidine were comparable in both products. Total Amino Acids (TAA) was significantly higher ($p<0.05$) in sorghum/cowpea with 993 than maize/cowpea *ogi* with 879.Ratio of total essential amino acids to total amino acids (TEAA/TAA) was higher in sorghum/cowpea with 0.43 than maize/cowpea *ogi* with 0.46. Total Sulphur Amino Acids (TSA). Amino Acid Scores (AAS) were comparable in both products but higher in maize/cowpea in the absence of tryptophan. Total aromatic amino acids values were higher in sorghum/cowpea with 9.2 than maize/cowpea.

DISCUSSION

High value of water absorption capacity is desirable for the improvement of mouth feel and viscosity reduction in food product. In this study, sorghum/cowpea *ogi* had a higher value (235%) than maize/cowpea *ogi* (103%); The WAC value of sorghum/cowpea ogi was higher than 130% reported for soy pea flour and lupin seed flour (Lin *et al*., 1974). This difference might be due to several factors like changes in quality and quantity of starch during fermentation process and starch- protein matrix network. There might also be likely exposure of charges which can attract the water molecules in the fermenting medium. The lower value of maize/cowpea *ogi* might be due to protein-protein interaction. The lower value of WAC in maize/cowpea is however is in agreement with the finding with of Ahmed *et al*. (1988) that reported that fermentation increase protein which was reflected in better WAC. Water absorption capacity of about 149.1-471.5% are considered critical in viscous foods and high viscous food is undesirable for infant complementary foods; thus the low water absorption capacity of co-fermented maize/cowpea might serve as a desirable product for complementary food; considering also that during boiling since the co-fermented mixture must be boiled before consumption, this might increase gelatinization and swelling thereby increasing water absorption potential. The oil emulsification capacity was higher in maize/cowpea *ogi* (15%) than that of sorghum/cowpea (5%) but the value of maize/cowpea was significantly higher than the reported value of 7-11% for wheat flour and 18% for soya flour (Lin *et al*., 1974). High value of oil emulsification capacity acts as flavour retainer and enhances the mouth feel and taste of food. Co-fermentation brings about variation in pH which might have contributed to the value in both products. The higher values of water absorption capacity and oil absorption capacity in sorghum/cowpea might be due to the thickness of interfacial bi- layer model of protein diffusion and re-orientation to water interface. The thickness of the interface also depends on protein-to-protein interaction. This justified why the higher value of crude protein in sorghum/cowpea gave a higher value of oil absorption capacity; this was in agreement with the report of Sefa-Dedeh and Afaokwa (2001).

The reduced value of oil absorption stability in maize/cowpea *ogi* might be due to collapse of proteins thereby increasing contact between protein molecules leading to coalescence and thus reducing stability.

Foaming Capacity

Low values of foaming capacity are indicative of soluble proteins and indicative of low gas/volume ratio. The value in maize/cowpea *ogi* was higher than sorghum/cowpea *ogi*. However, the value of maize/cowpea *ogi* (40%) in this study is comparable to the values reported for full fat cowpea flour (40%) (Abbey and Ibeh, 1988). The foaming stability values were favourably comparable in both products. The crude protein values in co-fermented sorghum/cowpea were comparable in both products (13.9 g/100 g) in sorghum/cowpea and maize/cowpea *ogi* (13 g/100 g). Least gelation capacity values of maize/cowpea and sorghum/cowpea *ogi* were low and comparable. High value of least gelation capacity means less thickening capacity of food product. Thus the low values in co-fermented mixtures might be appropriate for infant complementary foods.

Bulk Density

The values of packed bulk density were comparable (0.50-0.55 cm^3) were comparable in both samples. The values of bulk density of loose and packed flour of sorghum/cowpea *ogi* were comparable to that of maize/cowpea *ogi*. Thus it could be deduced that co-fermentation of maize or sorghum with cowpea might not increase the bulkiness of maize or sorghum *ogi*; high value of bulkiness is undesirable for infant complementary food due to the physiology of the alimentary canal and stomach capacity of the infant.

Proximate Composition

Higher values of dry matter might depend on the hydrophobic and hydrophillic nature of the cereals or cowpea.; the higher the dry matter, the higher the water retention capacity. Unfermented cereals/cowpea had higher dry matter than co-fermented cereal/cowpea *ogi* this might be due to the fact that unfermented grains were not soaked in water. This study showed that co-fermentation reduced crude protein of maize/cowpea and sorghum/cowpea *ogi* this finding is in agreement with Aliya and Geervani (1981) reported a reduction of the total crude protein content of 4-6% in fermented millet porridge. Akpapunam and SefahDedeh (1995) reported that there was no change in crude protein in fermented maize incorporated with cowpea in ratio 60:40 maize:Cowpea when compared with unfermented maize and cowpea mixtures. The values of crude protein in both co-fermented samples in this study were significantly ($p>0.05$) higher than estimated protein need (3.6 g day^{-1}) required from complementary food by level of usual breast milk intake for 6-23 month old infants; however the value in both samples were comparable to required protein recommended intake during the first two years of life (WHO/NUT, 1998). The low value of crude fibre of the two co-fermented samples in this work is desirable because the physiology of infants alimentary canal does not favour bulky foods. The reduction of crude fiber might also be due to enzymatic degradation of the fibrous material during fermentation (Ikenebomeh *et al*., 1986). According to the report of Akpapunam and Achinewu (1985), there was decrease in lipid content of pearl millet/cowpea blend of 60:40 ratios in fermentation process compared with raw blends. In this study, the lipid content of unfermented cereal/cowpea mixture was significantly higher ($p<0.05$) than their unfermented samples, while co-fermented sorghum/cowpea had a significantly higher value than co-fermented maize/cowpea.. The germ and the aleurone layers of millet and sorghum contribute lipid while the germ contribute about 80% of total fat. The presence of lipid in cereal starches is a distinguishing feature of the starches; in this study co-fermentation appeared to reduce the lipid content this is in agreement with the finding of Beuchat and Worthington (1974).

Reducing Sugars

Starch is the major storage form of carbohydrate in sorghum and maize. The digestibility of starch depends on hydrolysis of pancreatic enzymes. In this study, maize/cowpea had slightly lower reducing sugar content than sorghum/cowpea *ogi* (Table 1). Co-fermentation in this study, sharply reduced the reducing sugars in both samples, this is in agreement with Sripriya and Chandra (1997). The reduction might be due to hydrolysis of polysaccharides by fermenting microbes which possess both alpha and beta amylase; because carbohydrates, starch and soluble sugars (reducing sugars) are the principal substrates for fermenting lactics therefore significant decrease in soluble sugars is expected. The observation in this present work is however contrary to the finding of Chauvan (1988), who reported increase in the reducing sugar content during natural fermentation of sorghum for four days but reported decrease in reducing sugar when fermentation composition continued for seven days.

Amino Acids

The results showed that glutamic acid and leucine were the most abundant in both products. High value of glutamic acid might affect the flavour of the products and may cause ulcer in man. The high acids are products of microbial fermentation. The values of leucine of 129.3 mg g^{-1} cp in sorghum/cowpea and 101.1 mg g^{-1} CP in maize/cowpea were higher than the recommended dietary allowance (RDA) value of 93 mg g^{-1} cp for infant complementary food. There is amino acid antagonism within sorghum grain; leucine might depress isoleucine which with other factors is a contributor to pellargra disease resulting from consumption of sorghum. Isoleucine value was 35.9 mg g^{-1} cp in maize/cowpea and 38.2 mg g^{-1} cp in sorghum/cowpea both values were lower than RDA value of 46 mg g^{-1} cp. Arginine which is an essential amino acid for child's growth (Robinson, 1987) content was slightly higher in sorghum/cowpea 42.3 mg g^{-1} cp than maize/cowpea 36.3 mg g^{-1} cp. Arginine is an essential amino acid needed for child growth (Robinson, 1987). The lysine content of maize/cowpea *ogi* was slightly lower 22.8 than 42.3 mg g^{-1} cp of sorghum/cowpea *ogi*. However both values were lower than 63 mg g^{-1} of reference egg protein and RDA value (66 mg) for infant complementary food.

Methionine and cystine values were comparable in maize/cowpea 19.7 and 20.9 mg g^{-1} cp in sorghum/cowpea *ogi*. Methionine and cystine are considered the limiting amino acids in legumes. Methionine plus cystine values in both co-fermented mixtures were comparable with RDA values of 42 mg g^{-1} cp required for infant complementary foods. Methionine is needed for the synthesis of choline; which Choline forms lecithin when diet is low in protein resulting in Protein Energy Malnutrition (PEM). Sufficient choline may be formed in the presence of adequate methionine. Thus consumption of maize/cowpea or sorghum/cowpea *ogi* may solve methionine deficiency and also meet the RDA value. Cystine has positive effect on zinc ions and can act for part requirement of methionine, however, FAO/WHO/UNU(1985) does not give the proportion of Total Sulphur Amino Acid (TSAA) which can be met by cystine in man.

The histidine value was higher in sorghum/cowpea *ogi* than of maoze/cowpea *ogi*. Histidine is essential for infant growth when present in small quantities and when allergens enter the tissue histidine is liberated in large quantities and it is responsible for nettle rash (Adeyeye and Faleye, 2004). However the value of histidine in sorghum/cowpea was comparable to RDA while maize/cowpea *ogi* was lower to RDA value.

In the total amino acids maize/cowpea had a lower value of 879 mg g^{-1} cp than sorghum/cowpea of 993. Both values were however higher than that of 566 mg g^{-1} cp of egg reference protein reported by Paul *et al*. (1980) and 453 mg g^{-1} cp reported by Lusas (1979) for peanut meal. Both products appeared promising in satisfying the total amino acid requirements of infants. The Total Sulphur Amino Acids (TSAA) of sorghum/cowpea (42.2) and (39.5) for maize/cowpea; these values were lower than 58 mg g^{-1} cp recommended for infants (FAO/WHO/UNU, 1985). The aromatic amino acid (ArAA) of maize/cowpea 72.3 mg g^{-1} cp and 92.0 mg g^{-1} cp of sorghum/cowpea fell within the range suggested for ideal infant protein (68-118 mg g^{-1} cp) by FAO/WHO/UNU (1985).

The percent ratio of essential amino acid to the total amino acid in maize/cowpea and sorghum/cowpea were well above 26 and 11% recommended by FAO/WHO/UNU (1985) for ideal protein food for infants and adults, respectively. The values obtained for sorghum/cowpea and maize/cowpea were also comparable to that of egg (50%) (FAO/WHO/UNU, 1990).The values obtained for maize/cowpea also compared favourably with 43.6% reported for pigeon pea flour by Oshodi *et al*. (1993) and 43.8-44.4% reported by Chauvan *et al*. (2001) for beach pea protein isolate. The P-PER (Protein efficiency ratio) value of maize/cowpea was 3.9 g/100 g and sorghum/cowpea was 7.2 g/100 g both values were higher than literature values of (1.21) cowpea and (1.82) pigeon pea as reported by Salunkhe and Kadam (1989). The P-PER values were also higher than that of reference casein (2.50).

CONCLUSION

The higher protein quality in terms of amino acids composition of co-fermented sorghum/cowpea might have contributed to its higher WAC and OAC however, maize/cowpea had higher FC which was indicative of soluble proteins; in essence maize/cowpea might be more acceptable as complementary food. Also the lower WAC in maize/cowpea might make it to be more viscous, high viscous food is unacceptable as complementary food and its higher OEC would also make the maize/cowpea to be a good flavour retainer. However, both products had comparable low GC; the lower the value, the better, thus making both to have less thickening ability and to be good emulsifier. These attributes might make both products to be appropriate as complementary foods. The high value of glutamic and aspartic acid may affect the flavour. The values of leucine in both samples were higher than RDA levels while isoleucine and lysine were also lower than RDA. Although, However, histidine is needed for infants growth in small amounts. The value of histidine was lower in maize/cowpea than RDA level. The TAA values in both samples were higher in both samples than egg reference protein with sorghum/cowpea having higher value. Both products appeared promising in satisfying TAA requirements of infants and also ArAA infants' requirement. Crude protein values were comparable in both samples and meets the requirement for the first two years of life and higher than required level from complementary foods.

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