Preliminary study on the nutritional and functional properties of complementary food from malted millet (*pennisetum glaucum*) enriched with defatted and protein isolate of fluted pumpkin seed (*Teliferia occidentalis*)

ABSTRACT

The study evaluated the nutrient, antinutrient, mineral and functional properties of malted millet based complementary foods enriched with defatted and protein isolate from pumpkin. The millet were subjected to malting process while the pumpkin seeds were defatted and its protein isolated using a standard method. The proximate, antinutrient, mineral and functional properties of the complementary foods were analysed using standard analytical methods. The data obtained were analysed using descriptive statistic and Duncan’s multiple range test to determine the means and standard deviation at significant level of p< 0.05. The protein content ranged from 8.20 to 17.75 % while the ash and fat contents were 3.31 to 4.98 % and 2.56 to 7.65 %. The energy content was 358.33 to 422.96 Kcal. The tannin and phytate content are 0.15 to 0.50 mg/100 g and 0.55 to 0.96 mg/100 g while the saponin content was 0.30 to 0.69 mg/100 g. Calcium and Potassium content were 80.55 to 139.15 mg/100 g and 375.04 to 467.55 mg/100 g while sodium content was 45.87 to 101.24 mg/100 g. Water and oil absorption capacities were 60.44 to 88.92% and 75.00 to 99. 10 % while the bulk density was 0.55 to 0.82 g/ml. The swelling capacity and least gelation concentration were 60.0 to 96.87% and 12.00 to 20.22 %. The protein, fat and fibre content of the formulated complementary foods increased with addition of defatted and protein isolate of pumpkin. The micronutrient contents also showed a significant increase while the bulk density, swelling capacity and least gelation concentration also showed nutritional adequacy of the foods. Consequently, its application as a complementary food can make a difference in the onset and prevention of protein energy malnutrition among the infants in the developing countries.

Key words: Complementary foods, malted millet, protein isolate, protein energy Malnutrition, micronutrients.

INTRODUCTION

Breast milk is the most nutritionally balanced food that can be given to an infant during the first six months of life. It has all the right nutrients and immunological components that an infant needs to maintain a healthy living, promote growth and development. Breast milk also protects infants against the two leading causes of infant mortality which are upper respiratory infection and diarrhea (Issaka et al., 2015). However, after six months, breast milk alone will no longer be able to meet the nutritional demands of the growing infant whose weight is expected to have doubled. This is why complementary food needs to be introduced between the periods of 6 months to 24 months, to form part of the diet, when transition from exclusive breastfeeding to semi solid food is expected to commence. It is at this stage that the nutritional requirements of many infants are not met, thus leading to the onset of malnutrition that is prevalent in children under 5 years of age worldwide (Daelmans and Saadeh, 2003). This indeed has created
challenges for relevant stakeholders in the academia, food industry, government and foreign bodies to resolve on sourcing for locally developed and nutritious complementary foods that can be affordable by low income families or communities especially those in Sub-Saharan Africa. Nigeria is a country with an abundance of food that can be used for proper nutrition, as well as for the formulation of complementary foods (Lawan et al., 2018). A complementary food is any suitable food given to older infants and young children in addition to breast milk that provides additional nutrition to meet all growing child’s needs (Achidi et al., 2016; Fasuan et al., 2017). UNICEF (2018) also defines complementary food as “any non-breast milk foods or nutritive liquids that are readily consumed and digested by the young child and that provide additional nutrition to meet all the growing child’s needs during this period”. Most traditional weaning foods in developing countries are made from cereals, starchy fruits, root and tuber (Makinde and Lapido, 2012). They are known to be of low nutritive value and are characterized by low protein low energy and high bulk density (Makinde and Lapido, 2012). Cereal based diets have been implicated in protein-energy malnutrition (Mbaeyi-Nwaoha and Obetta, 2016). West Africa traditional weaning foods and feeding practices predispose the infants to malnutrition, growth retardation, infection and high mortality (Mbaeyi-Nwaoha and Obetta, 2016). Since many low income mothers are unable to afford most commercially available complementary foods, the use of locally developed, nutritious and affordable weaning foods from plant sources become imperative in order to address malnutrition among infants and very young growing children. Millets contain 7-11% proteins, 60-70% carbohydrates, 2-7% crude fibre and 2-5% fat. They are excellent source of vitamin B, magnesium, and antioxidants. Millet is also a good source of other dietary minerals like manganese, phosphorus and iron. Millet proteins are good source of essential amino acids except lysine and threonine but are relatively high in sulphur containing amino acids methionine and cysteine (Singh et al., 2012). Apart from this, some essential fatty acids like linoleic, oleic and palmitic acids found in free form and monogalactosyl, digalactosyl diacyl glycerols, phosphatidyl ethanolamine, phosphatidyl serine and phosphatidyl choline in the bound form present in millets (Bagdi et al., 2011). Fluted pumpkin (Telfairia occidentalis Hook, F) is regarded to be of high nutritional value in terms of protein, fat and minerals. The seed of fluted pumpkin, is popularly consumed in Nigeria, especially in the south eastern part of Nigeria where it is used as a condiment in soup. The fermented seeds of fluted pumpkin is used in the processing of “Ogiri ugu”, a locally made custard. The seeds of fluted pumpkin could also be used in cookie and marmalade manufacturing. The seed is also a good source of edible oil (Fagbemi, 2007). The combination of quality protein maize and defatted pumpkin has been explored as complementary food meant for children (Ikujenlola, 2013). However, the combination of millet, defatted and protein isolate from pumpkin has not been exploited as complementary food for the infants which can offer a good protein base diet that will satisfy the nutritional requirement of growing children thereby maintaining their health and development. Consequently, the objective of this research work is to produce high quality complementary food from blends of defatted, protein isolate of pumpkin flour and malted and unmalted millet and to assess the nutritional and functional characteristics of the diets.

MATERIALS AND METHODS

Materials

Millet grains used for this study was purchased from Sango market, Saki, Oyo State Nigeria, while the Pumpkin seeds were purchased from Oshodi market, Lagos State Nigeria.

Methods

Processing of unmalted millet flour

Raw millet grains were sorted, pretreated for 3 min with 200 ppm of bleach containing 5.25% sodium hypochlorite, and mixed in deionized water to control microbial growth as described by (IJarotimi et al., 2013). Seeds were rinsed, soaked in deionized water (1:3 w/v) for 5 h at ambient temperature (25-27°C), drained and ovendried at 45-55°C (Plus11, Sanyo Gallenkamp PLC, Leicestershire, UK) for 15 h. It was then milled using a Philips laboratory blender (HR2811 model) and sieved using a 60-mm mesh sieve (British Standard) to obtain raw millet flour. The flour was packed in a plastic container sealed with an aluminum foil and stored at room temperature (27°C).

Processing of malted millet flour

Millet grains were sorted, pretreated for 3 min with 200 ppm of bleach containing 5.25% sodium hypochlorite, and mixed in deionized water to control microbial growth as described by (IJarotimi et al., 2013). Seeds were rinsed, soaked in deionized water (1:3 w/v) for 5h at ambient temperature (25-27°C). Seeds were drained and placed on perforated aluminum pans lined with filter paper, then placed in temperature-controlled cabinet at 30°C for germination. After the fourth day, the germinating process was stopped. The germinated seeds were washed with distilled water and ovendried at 45-55°C (Plus11; Sanyo Gallenkamp PLC, Leicestershire, UK) for 15h. It was then milled using a Philips laboratory blender (HR2811 model) and sieved using a 60-mm mesh sieve (British Standard) to obtain malted millet flour. The flour was packed in a plastic container sealed with an aluminum foil and stored at room temperature (27°C).
Processing of defatted pumpkin seed

The modified method of Fagbemi (2007) was used for this purpose. The pumpkin seeds were washed, dried and milled using attrition mills. The resulting meal was defatted with food grade hexane as solvent. The defatted meal was oven dried at 45-55°C (Plus11; Sanyo Gallenkamp PLC, Leicestershire, UK) for 15h. It is then milled using a Philips laboratory blender (HR2811 model) and sieved using a 60-mm mesh sieve (British Standard) to obtain defatted pumpkin flour.

Processing of pumpkin protein isolate

Extraction of proteins and preparation of isolates was done using the method of Adebowale, et al. (2002) with slight modification by dispersing 1000g weight defatted pumpkin flour in distilled water in ratio 1:10. The mixture was stirred with a magnetic stirrer for 4h and pH of the solution was adjusted to 9.0 using 1 M HCl or NaOH and continued stirring for another 30 min at room temperature (25°C) to the point at which the protein was most soluble. Nonsolubilized materials were removed by centrifugation at 10,000 x g for 30 min. The residue was re-extracted two more times with the same solvent under similar conditions. The extracts were combined and proteins precipitated by adjusting the pH to 4.5 with 1 M HCl, followed by separation by centrifugation at 10,000 x g for 20 min. The precipitate was redispersed in 100 ml distilled water at pH 9.0 and reprecipitated at pH 4.5. After separation of proteins by centrifugation, the precipitate was washed twice with distilled water. The precipitated protein was re-suspended in distilled water and the pH was adjusted to 7.0 with 1M NaOH prior to freeze-drying. The freeze-dried protein isolates was stored in air-tight glass containers at room temperature.

Chemical analysis

Determination of proximate composition

The moisture, protein, crude fat, crude fibre and ash contents of the formulated malted millet-based complementary food were determined according to the standard methods of AOAC (2010). The Total Carbohydrate was determined by difference as described by (Akume et al., 2019):

\[ \text{Carbohydrate} = 100 - (\% \text{ moisture} + \% \text{ crude protein} + \% \text{ crude fat} + \% \text{ crude fiber} + \% \text{ total ash}) \]

Determination of the energy content

Energy content was determined using the method of Atwater factor as described by (Obiegbuna and Baba, 2001)

\[ \text{Energy (Kcal)} = (4 \times P) + (9 \times F) + (4 \times C). \]

Where:

- \( P \) = protein content (%)
- \( F \) = fat content (%)
- \( C \) = carbohydrate content (%)

Determination of the antinutritional content

Determination of tannin content: Tannin content was determined by the Folis-Denis colorimetric method described by Kirk and Sawyer (1998). 5 g of the sample was dispersed in 50 ml of distilled water and shaken. The mixture was allowed to stand for 30 min at 28°C, filtered through Whatman No 42 grade of filter paper. The extract (2 ml) was dispersed into a 50 ml volumetric flask. Also 2 ml standard tannin solution (tannic acid) and 2 ml distilled water were added in separate volumetric flasks followed by addition of reagent to each flask to serve as standard. Saturated 2.5 ml Na2CO3 solution was added. The content of each flask was made up to 50 ml with distilled water and allowed to incubate at 28°C for 90 min. Their respective absorbance was measured in a spectrophotometer at 260 nm using the reagent blank to calibrate the instrument at zero.

Determination of phytate content: Phytates was determined as described by Latta and Eskin (1980). 5 g of sample was added 50 ml of 0.8 M HCl solution for phytic acid extraction under orbital agitation (1300 rpm) for 1 h. The mixture was separated by centrifugation at 2800 rpm for 10 min. The supernatant eluted in ion-exchange chromatography column (0.50 g of Dowex-AGX-4 resin dissolved in 5 ml of deionized water) was used to prepare the stationary phase and 10 ml of deionized water was used for elution followed by 10 ml of 0.7M NaCl solution and again with 10 ml of deionized water. The supernatant 1 ml was diluted to 25 ml with deionized water, of which 2 ml was eluted in the column with 10 ml of 0.1 M NaCl solution, followed by 10 ml of 0.7M NaCl solution. The last aliquot was collected and 3 ml of the eluent was reacted with 1 ml of the Wade reagent (ferric chloride 0.03% and sulfosalicylic acid 0.5%). The absorbance was measured at 500 nm in a UV-visible spectrophotometer. The concentration of phytic acid was determined by difference as described by Onwuka (2005). About 2 g of the sample was suspended in 190 ml distilled water in a 250 ml volumetric flask, followed by the addition of 10 ml of 6M HCl at 100°C for 1h.
The digested sample was cooled and made up to 250 ml mark before filtration. Triplicate portion of 125 ml of the filtrate was measured into beakers and three drops of methyl red indicator was added. This was followed by the addition of conc. NH$_4$OH solution until the test solution changes from salmon pink color to a faint yellow color. Each portion was heated again to 90°C and 10 ml of 5% CaCl$_2$ solution was added while being stirred continuously. It was cooled and left overnight at 5°C. The solution was centrifuged at 2500 rpm for 5 min. The supernatant was decanted and the precipitate completely dissolved in 10 ml of 20% H$_2$SO$_4$ solution. The filtrate resulting from precipitation was made up to 300 ml. Filterate (25 ml) was heated until near boiling point and then titrated against 0.05 M standardized KMnO$_4$ solutions to faint pink color which persists for 30 sec.

**Determination of saponin:** Saponin was determined by the method described by Obadoni and Ochuko (2002). 20 g of each sample was added into a conical flask followed by the addition of 100 ml of 20% aqueous ethanol. The flask and its content was heated on a hot water bath for 4 h with constant stirring at 55°C filtered and the residue extracted further with 200 ml 20% ethanol. The combined extract was evaporated on a hot water bath at about 90°C until 40 ml volume was achieved. 20 ml Diethyl ether was added to the concentrate in 250 ml separator funnel followed by vigorous shaking. The aqueous layer was recovered while the ether layer was discarded. The purification process was repeated. 60 ml N-butanol was added and washed twice with 10 ml of 5% aqueous sodium chloride. The remaining solution was heated in a water bath after which the sample was dried in oven, weighed and saponin content was calculated as percentage.

**Determination of trypsin inhibitor:** The trypsin inhibitor activities (TIA) were determined using the procedure of (Kakade et al., 1974) which is based on the trypic hydrolysis of synthetic substrate, benzoyl-DL-arginine p-nitroanilide (BAPA). 1 g of finely ground and sieved samples of akee apple seeds and aril flour was defatted for 3 h using n-hexane. The samples were mixed with 50 ml of 0.01MNaOH and the pH was adjusted to 9.5 using 0.1M NaOH or 0.1MHC1. The mixture was macerated in warring blender for 2 min and centrifuged for 10 min at 1,000rpm. The extract from the sample was diluted with distilled water to obtain a dilution whereby 1 ml extract produced trypsin inhibitor activity of between 40-60%. Such dilution was used. The sample dilution was used with BAPA substrate and trypsin solution at 37°C. The reaction was allowed to take place in water bath for 10 min and their absorbance read at 410nm against the sample blank. Trypsin inhibitor activity (TIA) was calculated as:

\[
TIA = \frac{(2.632 \times D \times A1)}{S} = \text{mg pure trypsin/g sample}
\]

D = Dilution factor
A1 = Change in absorbance (pure trypsin and sample extract)
S = Sample mass.

**Mineral analysis**

The method described by Association of Official Analytical Chemists (AOAC, 2005) was used for mineral analysis. Two grams of each of the samples was digested with concentrated Nitric acid and Hydrogen peroxide, filtered and the filtrate in a 5 ml volumetric flask was loaded to Atomic Absorption Spectrophotometer, (model 703 Perkin Elmes, Norwalk, CT, USA). Calcium, magnesium, iron, sodium, potassium were determined at wavelengths 317.9 nm, 285.2 nm, 259.9 nm, 324.7 nm, and 213.9 nm respectively. Phosphorus was determined using Vanadomolybdate method. The serially diluted phosphate standard solution was made acidic by addition of 2 ml nitric acid (2:1), 25 ml of the Vanado-molybdate reagent was added. The solution was diluted to the mark with distilled water, mixed thoroughly and allowed to stand for 10 min and the optical density was measured at 470 nm. All values were expressed in mg/100g.

**Determination of functional properties of the diet**

**Determination of the bulk density**

Bulk density was determined according to the gravimetric method described by Mir et al. (2014). 10 g of sample was measured into a calibrated 50 ml measuring cylinder with repeated mild tapping, until a constant volume was observed. More samples were added to make up to the graduated line before measurement were taken and added it up to the graduated line before weighing. The results were reported as g/ml.

\[
\text{Bulk density (g/ml)} = \frac{\text{weight of sample (g)}}{\text{volume of sample (ml)}}
\]

**Determination of the water absorption capacity**

Water absorption capacity (WAC) was carried out according to the method described by Adebowale et al., (2005). 10 ml of distilled water was added to 1 g of the sample in a beaker. The suspension was agitated using magnetic stirrer for 3 min. The suspension obtained was thereafter centrifuged at 2,058 x g for 30 min and the supernatant was measured into a 10 ml graduated cylinder. The absorbed water by the flour was considered as the change between the initial volume of the water and the volume of the supernatant. The water density was taken as 1.0 g/ml.
\[ WAC = \frac{\text{weight of sample (g)}}{\text{volume of water used-volume of unabsorbed (ml)}} \times 100 \]

**Determination of oil absorption capacity**

Oil absorption capacity (OAC) which is an index of the amount of oil retained within a protein matrix under certain conditions was determined according to the method described by Adebowale et al. (2005). About 10 ml of oil known specific gravity was added to 1 g of sample in a beaker. The suspension was stirred using magnetic stirrer for 3 min. The suspension obtained was thereafter centrifuged at 3500 rpm for 30 min and the supernatant was measured into a 10 ml granulated cylinder. The density of oil used was 0.931 g/ml. The change between the original volume of the oil and the volume of the supernatant was calculated as the oil absorbed by the flour.

\[ \text{OAC} = \frac{\text{weight of sample (g)}}{\text{volume of water used-volume of unabsorbed (ml)}} \times 100 \]

**Determination of the swelling capacity**

Swelling capacity (SC) was determined using the modified method of Tosh and Yada (2010) with slight modification. 2.5g of the sample was measured in a 50 ml measuring cylinder. About 30 ml was added and mixed until homogeneity is reached. The mixture was then left to settle for 24 h, and the final volume \( V_f \) occupied by the sample was measured. The swelling capacity will be obtained as follows:

\[ \text{Swelling capacity (\%)} = \frac{V_f \text{ (ml)}}{\text{Sample weight (g)}} \times 100 \]

**Determination of foaming capacity**: The method of Jitngarmkusol et al., (2008) was used for the determination of the foaming capacity of the samples with some slight modifications. About 2 g of each flour sample was mixed with 100 ml of distilled water and the suspension was whipped with a kitchen blender. The whipped suspension was transferred into a 250 ml graduated cylinder. Volumes of the whole mixture were recorded before and after whipping and the experiment was done in triplicate. The foaming capacity was calculated using the equation:

\[ \text{Foaming capacity (\%)} = \frac{(V_1-V_3)/(V_2)}{V_3} \times 100 \]

\( V_1 \) is the volume of initial mixture
\( V_3 \) is the volume of the mixture after whipping and
\( V_3 \) is the volume of the foam after 5 h.

**Determination of least gelation concentration**

Test tubes containing 20% (w/v) dispersions of each diet were prepared with 5 ml distilled water. The dispersions were heated for 1 h in a boiling water bath, cooled rapidly under running tap water and subsequently at 4°C for 2 h. The test tubes were inverted to determine the concentration at which the sample would not slip.

**DATA ANALYSIS**

The software package used for the statistical analysis was the version 23 of the SPSS while all the analyses were carried out in three replicates and the standard error mean were calculated. The data were evaluated for significance differences \((p< 0.05)\) in their means using Analysis of Variance (ANOVA). Differences between means were separated using Duncan's Multiple Range Test.

**RESULTS AND DISCUSSION**

**Proximate composition of complementary foods from malted millet and pumpkin**

The moisture content of the complementary foods ranged from 7.56 to 8.75% with the malted millet-defatted pumpkin showed the highest moisture and the raw millet the lowest. The formulated complementary food has a higher moisture content when compared to 3.93 to 5.03% for complementary foods developed from the blends of sorghum-African yam beans-soybean reported by Bello et al., (2019). However, the values were within the range of 7.07 to 8.38% for complementary food made from malted quality protein maize and defatted fluted pumpkin flour (Ikujenlola et al., 2013). The high moisture of 8.35 and 8.75% observed in malted millet-defatted pumpkin and malted millet-pumpkin protein isolate may be due to the melting process of the millet in which more moisture was absorbed before malting was stopped.

The protein Advisory Group of the United Nations recommended that the moisture content of any floury products should not exceed 10% in order to extend the shelf life of such product, hence, the moisture content of the complementary foods fell within the recommended value. The malted millet-defatted pumpkin showed highest protein content of 17.75% while the raw millet have the lowest value of 8.20%. The protein value observed in this study were within the range of 8.33 to 17.60% for malted quality protein maize and defatted pumpkin complementary food reported by Ikujenlola et al., (2013) and 8.20 to 12.20% for maize-based complementary food supplemented with soybean and sweet potato flour (Okoye and Egbujie, 2018). The inclusion of defatted and protein isolate of pumpkin which is of high protein quality in terms of essential amino acids significantly increase the protein content of the formulated complementary foods. Malting of the millet might also improve the protein quality of the sample as Inyang and Zakari (2008) reported that there is always increase in protein during malting process as a
result of net synthesis of the enzymes of the enzymatic protein by germinating seeds. The ash content of the malted millet-defatted pumpkin complementary food has the highest value of 4.98% which was significantly (p< 0.05) differed from 3.31% for raw millet. The malted millet-pumpkin protein isolate complementary food also showed a high value of ash which was 4.90%. The ash content observed in this study was higher than 0.66-1.44% for maize-based complementary food supplemented with garden pea flour (Ukenyima et al., 2019) and 1.11-1.46% for complementary food from malted millet, plantain and soybean blends as reported by Bolarinwa et al., (2016). Moreover, the ash content in this study fell within the range of <5% recommended by codex for ash content that must be present in complementary food for the infants. The crude fat content ranged from 2.65 to 7.65% for raw millet and malted millet-defatted pumpkin complementary foods. The value obtained for fat content was higher than 3.31-3.47% for complementary food made from sprouted and unsprouted sorghum, and Irish potato and groundnut (Adegbanke et al., 2017). The increase in the fat content of the samples may be due to inclusion of defatted and protein isolate of the pumpkin as the seeds contains high amount of oil. The fibre content of the sample ranged from 2.35 to 2.93% for raw millet and malted millet-defatted pumpkin complementary foods. The fibre content for the raw millet-defatted pumpkin and malted millet-pumpkin protein isolate were 2.65 and 2.80% respectively. These values were higher than 1.85-2.37% for fermented maize-based complementary foods enriched with mungbean malt (Eucharia et al., 2020). The high amount of fibre observed in this study may due to high amount of fibre present in millet (2.80-3.20%) as reported by Ali et al., (2003). The carbohydrate content of the samples ranged from 58.34 to 91.78% for malted millet-defatted pumpkin and raw millet. The values for carbohydrate in this study were similar to 67.59-78.02% for complementary food made from sorghum, African yam bean and mango mesocarp flour blends (Yusufu et al., 2013). The relatively high carbohydrate is an indication that the formulated complementary foods would provide the infants with the adequate amount of energy. The energy content of the complementary foods decreased with the addition of both the defatted and pumpkin protein isolate as raw millet complementary food showed a value of 371.55 and 373.21 Kcal respectively as shown in Table 1. This value was similar to values reported by previous researchers (Anuonye, 2012; Itagi and Singh, 2012) in cereal-legumes mix complementary foods. The decrease in the energy values of the pumpkin substituted complementary foods could be due to low fat and carbohydrate of the blends as both constitute major source of energy.

**Table 1:** Proximate composition (%) and energy (Kcal) of complementary food from malted millet and pumpkin.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Moisture</th>
<th>Protein</th>
<th>Ash</th>
<th>Fat</th>
<th>Fibre</th>
<th>CHO</th>
<th>Energy</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw millet</td>
<td>7.56±0.01</td>
<td>8.20±0.01</td>
<td>3.31±0.02</td>
<td>2.56±0.01</td>
<td>2.35±0.02</td>
<td>91.78±0.01</td>
<td>422.96±0.02</td>
</tr>
<tr>
<td>Malted millet</td>
<td>7.78±0.01</td>
<td>10.35±0.01</td>
<td>4.50±0.02</td>
<td>3.45±0.01</td>
<td>2.45±0.01</td>
<td>71.47±0.02</td>
<td>358.33±0.02</td>
</tr>
<tr>
<td>Raw millet + pumpkin protein isolate</td>
<td>7.66±0.02</td>
<td>13.34±0.02</td>
<td>4.65±0.02</td>
<td>4.45±0.01</td>
<td>2.55±0.02</td>
<td>67.35±0.02</td>
<td>362.81±0.01</td>
</tr>
<tr>
<td>Raw millet + defatted pumpkin</td>
<td>7.69±0.01</td>
<td>15.67±0.01</td>
<td>4.87±0.01</td>
<td>5.65±0.02</td>
<td>2.65±0.02</td>
<td>63.47±0.01</td>
<td>367.41±0.02</td>
</tr>
<tr>
<td>Malted millet + pumpkin protein isolate</td>
<td>8.75±0.01</td>
<td>16.76±0.02</td>
<td>4.90±0.01</td>
<td>6.67±0.02</td>
<td>2.80±0.01</td>
<td>61.12±0.02</td>
<td>371.55±0.01</td>
</tr>
<tr>
<td>Malted millet+ defatted pumpkin</td>
<td>8.35±0.02</td>
<td>17.75±0.01</td>
<td>4.98±0.01</td>
<td>7.65±0.02</td>
<td>2.93±0.02</td>
<td>58.34±0.01</td>
<td>373.21±0.01</td>
</tr>
</tbody>
</table>

Values with different subscripts in a column are significantly (p<0.05) different.

Antinutrient composition of complementary foods from malted millet and pumpkin

The antinutrient composition of the complementary foods is shown in Table 2. The tannin, phytate and
oxalate contents of the samples ranged from 0.15 to 0.50 mg/100 g, 0.55 to 0.96 mg/100 g and 0.09 to 0.28 mg/100 g for malted millet- defatted pumpkin and raw millet respectively. It could be observed from this study that malting of the millet significantly decreased the tannin, phytate and oxalate content when compared with unmalted millet and this could be as a result of formation of hydrophobic association of tannin, phytate and oxalate during malting. It could also be due to leaching of tannin, phytate and oxalate into the water during the malting process (Afam et al., 2016). The tannins content of the formulated complementary foods in this study was within the range of 2.55-2.85 mg/100 g recommended for infant complementary foods as reported by (Onaja et al., 2014). Vasumdhara et al., (2013) reported that an excess of 800 mg/100 g phytate per day is not a good idea. It has also been reported that phytate is not readily absorbed by human and that it affects mineral bioavailability only in combination with mineral poor diets. Tannins are polyphenolic compounds found in many plants. They often lower the absorption of some materials into the body most especially iron absorption (Ashok and Upadhyaya, 2012). Phytate is found in legumes, nuts and oil seeds and is associated with proteins and often associated with protein absorption in the body and they are often isolated or concentrated with protein fraction of these food materials (Khokhar and Owusu, 2003). Soaking, cooking, germination/malting and roasting has been found to reduce the level of antinutritional factors in food, thereby enhancing the bioavailability of the nutrients in foods (Kumar et al., 2010). Consequently, the phytate level of the formulated foods are within the safe level and will not pose any health challenge. Saponnin and trypsin inhibitor of the samples ranged from 0.30-0.69 mg/100 g and 0.75 to 1.69 mg/100 g for malted millet-defatted pumpkin and raw millet complementary foods respectively. The saponnin value observed in this study was lower to the value of 1.41-3.13 mg/100 g for complementary food from malted millet, plantain and soybean blends (Bolarinwa et al., 2016). Saponnin are water-soluble glycosides in which the non-sugar moiety is a steroid. They are gastric irritants but may also exhibit hypocholesterolemic activity, however at larger quantities, they have toxic properties causing haemolysis of the red blood cells. (Pulkkinen et al., 2015).

### Table 2: Antinutrient composition (mg/100 g) of complementary food from malted millet and pumpkin.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Tannin</th>
<th>Phytate</th>
<th>Oxalate</th>
<th>Saponin</th>
<th>Trypsin inhibitor</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw millet</td>
<td>0.50±0.01</td>
<td>0.96±0.01</td>
<td>0.28±0.02</td>
<td>0.69±0.02</td>
<td>1.69±0.01</td>
</tr>
<tr>
<td>Malted millet</td>
<td>0.40±0.01</td>
<td>0.80±0.01</td>
<td>0.25±0.02</td>
<td>0.58±0.02</td>
<td>1.32±0.02</td>
</tr>
<tr>
<td>Raw millet + pumpkin protein isolate</td>
<td>0.33±0.02</td>
<td>0.80±0.02</td>
<td>0.20±0.01</td>
<td>0.46±0.01</td>
<td>1.15±0.01</td>
</tr>
<tr>
<td>Raw millet + defatted pumpkin</td>
<td>0.30±0.01</td>
<td>0.70±0.02</td>
<td>0.15±0.01</td>
<td>0.40±0.02</td>
<td>1.10±0.01</td>
</tr>
<tr>
<td>Malted millet + pumpkin protein isolate</td>
<td>0.20±0.01</td>
<td>0.68±0.02</td>
<td>0.10±0.01</td>
<td>0.36±0.01</td>
<td>0.90±0.01</td>
</tr>
<tr>
<td>Malted millet+ defatted pumpkin</td>
<td>0.15±0.02</td>
<td>0.55±0.02</td>
<td>0.09±0.02</td>
<td>0.30±0.01</td>
<td>0.75±0.02</td>
</tr>
</tbody>
</table>

Values with different subscripts in a column are significantly (p<0.05) different.

### Mineral composition of complementary foods from malted millet and pumpkin

The calcium and iron content of the formulated complementary food ranged from 80.55 to 139.15 mg/100 g and 6.50 to 8.99 mg/100 g as shown in Table 3. There was significant (p<0.05) difference in the calcium and iron content of the samples. The result for these minerals showed an increasing trend with addition of defatted protein isolate of pumpkin. The calcium content was higher than the calcium content of 6.44-12.14 mg/100 g of complementary gruel from sorghum, soybean and plantain (Onoja et al., 2014) but fell within the range of 91.00-121.33 mg/100 g for complementary foods from sorghum, irish potato and groundnut (Adegbanke et al., 2017). The iron content in this study were higher than 2.99-4.38 mg/100 g for sorghum-African yam bean complementary food (Okoye et al., 2017). Higher calcium and iron level reported in the malted millet-defatted pumpkin and malted millet-pumpkin protein isolate could be due to the influence of malting on the millet grains as malting has been reported to increase in vitro extractability and bioaccessibility of minerals like calcium, iron and Zinc (Suma and Uroji, 2011; Krishnan et al., 2012). Calcium is an essential nutrient in the mineralization of bones and teeth and for regulating intracellular activity in the body tissue (Deborah, 2007). Iron on the other hand is essential for the formation of the blood cells and prevention of anemia in infants and children. The potassium, sodium and phosphorus contents ranged from 360.55 to 467.55...
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Table 3: Mineral compositions (mg/100 g) of complementary food from malted millet and pumpkin.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Calcium</th>
<th>Iron</th>
<th>Potassium</th>
<th>Sodium</th>
<th>Phosphorus</th>
<th>Magnesium</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw millet</td>
<td>80.55±0.01</td>
<td>8.75±0.02</td>
<td>467.55±0.02</td>
<td>45.87±0.01</td>
<td>250.11±0.01</td>
<td>15.55±0.02</td>
</tr>
<tr>
<td>Malted millet</td>
<td>95.88±0.01</td>
<td>6.65±0.01</td>
<td>385.45±0.01</td>
<td>85.47±0.01</td>
<td>275.55±0.02</td>
<td>13.25±0.01</td>
</tr>
<tr>
<td>Raw millet + pumpkin protein isolate</td>
<td>101.55±0.02</td>
<td>6.50±0.01</td>
<td>380.56±0.02</td>
<td>80.44±0.02</td>
<td>261.12±0.02</td>
<td>14.14±0.01</td>
</tr>
<tr>
<td>Raw millet + defatted pumpkin</td>
<td>120.11±0.01</td>
<td>7.70±0.02</td>
<td>360.55±0.01</td>
<td>82.77±0.01</td>
<td>270.11±0.01</td>
<td>15.75±0.01</td>
</tr>
<tr>
<td>Malted millet + pumpkin protein isolate</td>
<td>130.12±0.02</td>
<td>8.50±0.01</td>
<td>375.04±0.02</td>
<td>89.44±0.02</td>
<td>280.25±0.01</td>
<td>15.75±0.01</td>
</tr>
<tr>
<td>Germinated millet + defatted pumpkin</td>
<td>139.15±0.01</td>
<td>8.99±0.02</td>
<td>385.66±0.01</td>
<td>101.24±0.01</td>
<td>285.72±0.02</td>
<td>16.83±0.01</td>
</tr>
</tbody>
</table>

Values with different subscripts in a column are significantly (p<0.05) different.

Table 4: Functional properties of complementary food from malted millet and pumpkin.

<table>
<thead>
<tr>
<th>Samples</th>
<th>Bulk density (g/ml)</th>
<th>Foam capacity (%)</th>
<th>Least gelation concentration (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Raw millet</td>
<td>0.82±0.01</td>
<td>4.50±0.01</td>
<td>12.00±0.01</td>
</tr>
<tr>
<td>Malted millet</td>
<td>0.75±0.02</td>
<td>3.10±0.02</td>
<td>14.00±0.01</td>
</tr>
<tr>
<td>Raw millet + pumpkin protein isolate</td>
<td>0.70±0.02</td>
<td>3.15±0.01</td>
<td>16.60±0.02</td>
</tr>
<tr>
<td>Raw millet + defatted pumpkin</td>
<td>0.65±0.02</td>
<td>5.00±0.02</td>
<td>17.76±0.02</td>
</tr>
<tr>
<td>Malted millet + pumpkin protein isolate</td>
<td>0.62±0.01</td>
<td>4.60±0.02</td>
<td>18.64±0.01</td>
</tr>
<tr>
<td>Malted millet + defatted pumpkin</td>
<td>0.55±0.02</td>
<td>4.20±0.02</td>
<td>20.22±0.02</td>
</tr>
</tbody>
</table>

Values with different subscripts in a column are significantly (p<0.05) different.

mg/100 g, 45.87 to 101.24 mg/100 g and 250.11 to 285.72 mg/100 g respectively. Potassium content observed in this study was lower to 436.00-550.00 mg/100 g for complementary from mixtures of malted quality protein maize and steamed cowpea (Ikujenlola and Adeturoye, 2014). The sodium content of the food was higher than 2.80-3.18 mg/100 g for complementary foods from malted millet, plantain and soybeans blends (Bolarinwa et al., 2016). The Phosphorus content was lower to 322.18-355.85 mg/100 g for complementary from mixtures of malted quality protein maize and steamed cowpea (Ikujenlola, 2014). Potassium helps in blood clotting and in proper functioning of the muscles. Sodium is an essential cation in extracellular fluid in the body and is an important nutrient necessary for the maintenance of plasma volume, acid-base balance and normal cell function (Verbalis et al., 2010). Phosphorus is an important constituent of every living cell and also very essential in bone formation and other cellular reactions in the body (Berdanier and Zemplen, 2009). The magnesium content of the sample ranged from 13.25 to 16.83 mg/100 g with the malted millet-defatted pumpkin having the highest value and the malted millet the lowest. Malted millet-pumpkin protein isolate also showed a high value of Magnesium (15.75 mg/100 g). The addition of defatted pumpkin and its protein isolate improves the magnesium content of the complementary foods. The obtained values from this study was higher than 3.47-4.15 mg/100 g for maize-based complementary food fortified with soybean and sweet potato flour (Okoye and Egbujie, 2018). Magnesium helps in the proper functioning of the muscles.

Functional properties of complementary foods from malted millet and pumpkin

The bulk density of the formulated complementary foods is shown in Table 4. The value ranged from 0.55 to 0.82 g/ml.
for malted millet-defatted pumpkin and raw millet complementary foods respectively. The bulk density of sample in this study fell within the range of 0.72-0.80 g/ml for complementary foods from teff fortified with soybean and orange-fleshed sweet potato (Tenagashaw et al., 2016). However, it was found to be higher than the value of 0.17-0.29 g/ml for breakfast cereals from maize, African yam bean and coconut (Usman, 2012). The malting of the millet grains resulted in the low bulk density observed in malted millet-defatted pumpkin complementary food. Malting has been reported to be useful in preparation of low bulk density complementary food as low density complementary foods will not limit the calorie intake per feed for the infants and they will also be able to consume enough to satisfy energy and dietary intake requirement (Okoye et al., 2010; Omueti et al., 2009). The foam capacity of the sample ranged from 3.15 to 5.00 % for malted millet and malted millet-pumpkin protein isolate. The result obtained fell within the range of 2.83 to 7.69 % for weaning food processed from maize, pawpaw, red beans and mackerel fish meal (Bernard et al., 2016). Malting was found to reduce the foaming capacity of the formulated complementary foods. Malting process has been reported to have a decreasing effect on foam capacity of complementary foods (Okafor and Usman, 2014). Foam capacity is used to determine the ability of flour to foam which is dependent on the presence of flexible protein molecules which decrease the surface tension of water (Ohizua et al., 2017). The least gelation concentration is the minimum amount of flour needed to form a gel in a measure volume of water (Ohizua et al., 2017). The least gelation concentration of the samples ranged from 12.00 to 20.22 %. It is shown from this result that the raw millet gels at lower percentage while the malted millet-defatted pumpkin gels at higher percentage. Gels are characterize by their viscosity, plasticity and elasticity, hence the higher the least gelation concentration, the lower is the ability of the flour to form a stable gel (Lawan et al., 2018). It could therefore be deduced that raw millet diet will form a stable gel than the malted millet-defatted pumpkin and malted millet-pumpkin protein isolate. Flour product that form a stable gel could serve as a good binder and provide consistency in food preparation such as semi-solid beverages like kunu-zaki (Msheliza et al., 2018). Consequently, high least gelation concentration observed in malted millet-defatted pumpkin and malted millet-pumpkin protein isolate are also desirable as Adeoti and Osundahunsi (2017) reported that high least gelation concentration will lead to reduction in viscosity which results to increase in nutrient density and low bulk dietary bulk. The water and oil absorption capacities of the complementary foods ranged from 60.44 to 88.72 % and 75.00 to 99.10 % respectively as shown in Figure 1.

The values obtained for water and oil absorption capacities in this study were significantly lower than 135.00-165.00 % and 142.00-160.00 for complementary foods from sorghum-maize-mungbean (Onwurafor et al., 2017). Malting of the millet caused a decrease in the water and oil absorption capacities of the samples. The observed reduction was due to the fact that amylase enzyme activities developed during the malting process as these enzymes degraded the starch which is a major constituent of gel structure resulting in the production of a liquid gruel. These parameter is used parameter is used in determining the quantity of water the flour can absorbed and the degree of swelling within a particular time (Ayo et al., 2011; Marta and Tensiska, 2017). Complementary foods are expected to have a low water absorption capacity in order to produce a
more nutritious food (Mbatha et al., 2009), therefore, the low water absorption capacity of malted millet-pumpkin protein isolate and malted millet defatted pumpkin would be suitable complementary food for the infants. Oil absorption capacity is an important functional properties in food formulation as it enhances mouthfeel while retaining the food flavor (Obasi et al., 2018). It has been reported by Desalegn et al., (2015) that high oil absorption capacity is important for increasing energy density of complementary foods. It could therefore be concluded that malted millet-pumpkin protein isolate and malted millet-defatted pumpkin which has a high oil absorption capacity will increase the energy density of the formulated complementary foods. The swelling capacity of the complementary foods ranged from 60.00 to 96.87 %. This value was lower than 181.88 to 189.88 % for complementary foods from blends of sprouted paddy rice, sprouted African yam beans and pawpaw fruit. Swelling capacity of a sample illustrates the ability of the sample to absorb a particular amount of water and some within the duration of study (Fikiru et al., 2017).

CONCLUSION
The study showed that the inclusion of defatted pumpkin into malted millet generally increased both the macronutrients and micronutrients of the formulated complementary foods as high protein, fat and ash, calcium, Iron and potassium contents were observed, hence the complementary foods can be used by the infants to quantitatively and qualitatively enhance their nutritional status by mitigating the prevalence of protein energy malnutrition among the infants in developing countries.

REFERENCES