

Chemical Composition of Fortified, Ready-to-Use Maize-Bambara Groundnut Malt and Maize-Cowpea Malt Complementary Foods for Boosting Immunity in Infants Against Flu-Like Diseases

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Abstract

Maize-bambara groundnut malt and maize-cowpea malt complementary foods were fortified with a flour blend of edible termite and *Hibiscus sabderiffa* calyces, fermented and subjected to extrusion cooking. The fortified flour blends were evaluated for proximate composition, immune boosting micronutrients and chemical compounds known to be effective against flu-like viruses. The protein, carbohydrate, moisture, lipid, ash and crude fiber contents were 14.78-32.69, 48.05-70.47, 3.60-4.58, 3.33-12.00, 2.60-5.28, 1.17-1.80 percent respectively. The tannin, phytate and oxalate contents in mg/100g were 0.272-0.644; 0.193-0.938 and 1.114-1.575 respectively. The phenolic, flavonoid and alkaloid contents in μ g/g were 2240.26-2907.85; 167.22-566.43 and 0.33-2.47 respectively. Calcium was at a level of 0.55-4.99 mg/g, iron (1.71-7.32 mg/g), zinc (0.71-1.82 mg/g), selenium (0.004-0.390 μ g/g), vitamin A (1.15-2.99 μ gRE/g) and vitamin D (0.003 to 0.010 μ g/g). Of these chemicals, selenium and vitamin D have been reported to be immune boosters against flu-like diseases, but only the selenium content (0.21-0.39 μ g/g) met the RDA for infants aged 6-24 months with the fortified and extruded maize-bambara groundnut malt having the highest selenium content.

Keywords: Maize-cowpea malt, Maize-bambara grountnut malt, edible termite, *Hibiscus sabdariffa*, fortification, extrusion, immune boosting chemicals

Introduction

Breastfeeding provides the ideal food necessary for infants in the first-six months, after which the infants' requirements can no longer be satisfied by breast milk alone. At this point, it becomes necessary to supplement breast milk with high nutrient-containing adult foods to satisfy the nutritional needs of the infant for optimal growth and development (WHO, 2021). These nutritional requirements are further complicated during disease conditions especially when the immune system of infants is inadequately developed and vaccination is not an option. During such conditions, infants need more protein of good quality and micro-nutrients for normal physiological functioning, growth and maintenance (Ahmed et al., 2013) and to boost their Immunity against diseases.

Traditional complementary foods and cereal-legume blends designed to improve the quality of the traditional complementary foods in developing countries (Attaugwu and Uvere, 2021; Abesha et al., 2016) do not satisfy these needs while factory-manufactured complementary foods are designed to meet the basic metabolic needs of infants.

The COVID-19 pandemic claimed several lives, according to the report of Flaxman et al. (2023), about 821 deaths occurred as a result of COVID-19 in children and the young people aged 0-19 years in USA. Attempts to combat the virus by the use of drugs, vaccines, traditional herbs and medicines (Lim et al., 2021) have had some encouraging results. There are indications that food components such as lauric acid,

selenium, zinc, vitamins A, C, D and E could contribute to boosting the immune system against the COVID-19 virus (Malochleb, 2020; IFTNEXT, 2020). The use of these food components has however, not been explored exhaustively, particularly for weanlings during flu-like pandemics such as COVID-19.

This is the basis for this investigation aimed at producing and evaluating maize-bambara groundnut malt and maize-cowpea malt complementary foods fortified with the nutritionally dense and delicious edible termite (Kinyuru et al., 2013, Ezeocha et al., 2020) and phytochemical and micronutrient-rich *Hibiscus sabdariffa* calyces (Attaugwu and Uvere 2017a). This would help to tackle poor diet problems which is a major risk factor for a range of chronic diseases such as flu which result from poor immune system (Gombart et al., 2020). Hence, the main objective of this study was to evaluate the chemical composition of complementary foods produced from fermented maize-bambara groundnut malt and maize-cowpea malt blends fortified with edible termite and *Hibiscus sabdariffa* calyces using extrusion cooking to make it ready-to-use.

Methods

Materials

Processing of Maize, Bambara Groundnut and Cowpea for Production of Complementary Food

Flours from de-germed maize, malts of bambara groundnut and cowpea, ashed *H. sabdariffa* were produced by methods described by Attaugwu and Uvere (2021). Flours from the edible termites (*Macrotermes bellicosus*) was produced as described by Kinyuru et al., (2013). The ratio of the maize to bambara/cowpea malt in the composite flour was 70:30. The ratio of the termite to ashed Hibiscus calyces in the fortificant mix was 1.81:1 while the ratio of the composite flours to the fortificants was 10.56:1. The fortificants were mixed with the composite flour blends and fermented by backslopping for 72 hours (Nout et al., 1989). The samples were then oven dried for an hour at 100°C using a hot air Gallenkamp oven (Model IH, Gallenkamp, England), after which the cake was milled, sieved (0.5 mm sieve), and the flour stored in polythene bags at 10°C in a refrigerator. The extrusion cooking process was achieved using a pilot scale co-rotating twin screw food extruder (model Clextral, BC-21 N0 194, Firminy, France). The extruder was operated at a barrel temperature of 110°C with the feed moisture content in the extruder barrel at 18% (180 g/kg). The extruder feed rate was 5.1 g/min at a screw speed of 200 rpm. The extrudate was allowed to cool to 20°C, and then dried in a convective air oven at 105°C for 30 minutes, cooled, milled, sieved (0.5 mm sieve), and the flour stored in polythene bags at 10°C in a refrigerator.

Techniques

Diastatic Activity

The diastatic activities of the bambara groundnut and cowpea malts were determined by the method of Hulse et al., (1980) as modified by Uvere et al. (2010).

The fortified complementary foods, hibiscus calyces and termites were analyzed for proximate composition using the methods described by the A.O.A.C, (2010). Calcium Iron, Selenium and Zinc were determined using the Atomic Absorption spectrophotometric method as described by Norhaizan and Nor Faizadatul (2009). The vitamin A content was determined by the method described by Biesalski, et al., (1986). The tannin content was determined by the Folin-Denis spectrophotometric method as described by Pearson, (1976). Oxalate content was determined using the spectrophotometric method of Fassett (1973). The phytic acid content was determined by the method described by Lolas and Markakis (1975). The total flavonoids content was determined by the method described by Agbo et al., (2015). The phenol content was determined by the method described by Aadil, et al., (2013).

Statistical Analysis

Data analysis was carried out using one-way analysis of variance (ANOVA) in a completely randomized design (CRD); mean separation was by Duncan's New Multiple Range Test (Steel and Torrie, 1980). Significance was accepted at 5% level of difference.

Results

Malting characteristics of bambara groundnut and cowpea

Root length

The root length of the germinating legumes is presented in Fig.1. The root length increased with germination time and ranged from 0.93 cm to 3.05 cm and 0.33 to 1.76 cm for bambara groundnut and cowpea respectively. In bambara groundnut, root outgrowth was evident within the first 48 hours while in cowpea, root outgrowth was noticeable within the first 24 hours.

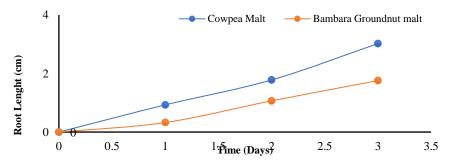


Figure 1: Root Length of Germinating Bambara groundnut and Cowpea Seeds

Malting Loss and Diastatic Activity

The malting loss and diastatic activity of the legume flours after 72 hours of malting is presented in Table 1. The malting loss was 18% in bambara groundnut malt and 16.55% in cowpea malt; the diastatic activity of the bambara groundnut malt and cowpea malt increased from 44.5 to 45.20°L and 45.50 to 45.67°L respectively after 72 hours of malting. This represents increases of 1.57% and 0.37% in the bambara groundnut malt and cowpea malt respectively.

Table 1: Malting Characteristics of Bambara Groundnut and Cowpea seeds

Samples	Root length increase	Malting yield	Malting loss (%)	Diastatic activity (°L)
Cowpea malt	66.67 ± 0.01	83.45 ± 0.01	16.55 ± 0.01	45.67 ± 0.23
Bambara groundnut malt	72.22 ± 0.01	82.00 ± 0.01	18.00 ± 0.01	45.20 ± 0.87

Proximate Composition of the Fortified Complementary Food Blends.

The proximate composition of extruded maize-bambara groundnut and maize-cowpea malt complementary foods are presented in Table 2.

The **moisture content** of the complementary food blends ranged from 3.62-4.58g/100g with sample (MB_{mf})_{df} having the highest moisture content of 4.58 g/100g while sample MC_m had the least value of 3.60 g/100g. The extruded complementary foods had moisture content ranging from 3.60 – 3.86 g/100g while the moisture content of fermented and dried complementary foods ranged from 4.07 - 4.58/100g. There was a significant (p<0.05) decrease in the moisture content of the formulated complementary foods after extrusion cooking. The fat content of the complementary foods ranged from 3.33 - 12.00 g/100g with sample (MC_{mf})_{df} having the highest fat content (12.00 g/100g) while sample MB_m had the least value (3.33 g/100g). There was a significant increase in the fat content of the complementary foods with the inclusion of the fortificants. The **crude protein** content of the complementary foods ranged from 14.78 - 32.69 g/100g with sample MC_{mf} having the highest protein content; the fortified samples [(MB_{mf})_{df.} (MC_{mf})_{df.} MB_{mf.} MC_{mf.}] had higher protein content ranging from 25-32.69 g/100g compared to the proprietary formula with a protein content of 15.0 g/100g. The fermented and dried complementary foods had a significant (p < 0.05) increase in crude protein with the inclusion of the fortificants. The formulated complementary foods showed higher ash content (2.60 - 5.28 g/100g) than the proprietary formula (2.50 g/100g). The **crude fiber** of the complementary foods ranged from 1.17 – 1.89 g/100g with (MC_{mf})_{df} having the highest value and MB_m the lowest.

There was a decrease in the carbohydrate content of the fermented and dried samples on inclusion of the fortificants and ranged from 48.05 - 70.47 g/100g.

Calcium, Iron, Zinc, Selenium, Vitamins A and D contents of the Maize-Bambara groundnut malt and Maize-Cowpea malt Complementary Foods.

The mineral and vitamin composition of the complementary foods are presented in Table 3.

The **calcium** content of the samples ranged from 0.55 - 4.99 mg/g with samples (MC_m)_{df} having the lowest (0.55 mg/g) and MB_{mf} having the highest (4.99 mg/g) values. The proprietary formula contained a calcium content of 3.50 mg/g which was lower than that of samples (MB_{mf})_{df} (4.10 mg/g), (MC_{mf})_{df} (3.85 mg/g), (MB_{mf}) (4.99 mg/g) and (MC_{mf}) (4.12 mg/g) complementary foods. There was a significant increase in the calcium content of the fortified fermented and dried complementary foods as compared to the unfortified blends. The **iron** content of the complementary foods ranged from 1.71 to 7.32 mg/g. The fermented and dried samples ranged from 1.71 to 7.04 mg/g while that of the extruded complementary foods was 1.74 - 7.32 mg/g, representing 1.8 to 4.0% increase in iron content. The fortified samples had iron contents ranging from 5.40 to 7.32 mg/g with sample (MB_{mf})_{df} having the lowest while (MC_{mf}) had the highest value.

Table 2: Proximate Composition of Complementary Foods

Proximate Compositio	(MB _m) _{df}	$(MB_{mf})_{df}$	(MC _m) _{df}	(MC _{mf}) _{df}	MB _m	MB_{mf}	MCm	MC_{mf}	P		RDA 6-23		
n g/100g										6-11 mo	mo	12-23 mo	6-23 mo
Crude Protein	14.78°±0.09	25.96e±0.58	17.28 ^b ±0.03	29.02 ^f ±0.22	18.02°±0.42	28.82 ^f ±0.15	20.21 ^d ± 0.45	32.69g±0.14	15.00°±0.0	3-4.5	3-5.5	4-6.5	6-11
Ash	$2.97^\text{c} \pm 0.02$	$4.71^{\rm f}\pm0.14$	$2.60^\text{b} \pm 0.03$	$4.67^{\mathrm{f}} \pm 0.02$	$3.49^{\text{d}} \pm 0.02$	$5.28^{h}\!\!\pm0.02$	$3.85^\text{e} {\pm}~0.03$	$5.19^{\text{g}} \pm 0.01$	$2.50^{\mathtt{a}} \!\! \pm 0.00$			-	-
Fat	$6.55^a \pm 0.06$	$10.15^{b} \pm 0.21$	$7.10^c \pm 0.14$	$12.00^{d} \pm 0.03$	$3.33^e \pm 0.03$	7.25°± 0.06	5.41 ^f ± 0.01	$9.13^{\text{g}} \pm 0.04$	10.00 ^b ±0.0	4.20	6.30	8.20	12.70
Moisture	$4.08^{\text{a}} \pm 0.04$	$4.58^b \pm 0.01$	$4.07^{\text{a}} \pm 0.00$	$4.37^\text{b} \pm 0.01$	$3.78^\text{c} \pm 0.01$	$3.86^{\text{d}} {\pm}~0.01$	$3.62^\text{e}{\pm}~0.02$	$3.74^{\rm f}\pm0.02$	$3.00^{g} \pm 0.00$		-	-	-
Crude Fibre	$1.31^b \pm 0.01$	$1.78^a \pm 0.02$	$1.39^c \pm 0.02$	$1.89^d \pm 0.01$	$1.17^e \pm 0.03$	$1.53^{£}\pm0.03$	$1.22^{g} \pm 0.02$	$1.74^a \pm 0.01$	$4.50^{h} \pm 0.00$	-	-	-	-
Carbohydrat e	70.31°±0.23	52.82 ^a ±0.21	67.36 ^b ±0.14	48.05 ^d ±0.46	70.47°±0.05	53.26 ^a ±0.03	67.69b±0.70	49.51°±0.02	65.00 ^f ±0.05	-	-	-	-

MBm - maize-bambara groundnut malt; MB_{mf} : fortified and extruded maize-bambara groundnut; MC_{m} : extruded maize-cowpea malt; $(MB_m)_{df}$: fermented and dried maize-bambara groundnut malt; $(MB_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-cowpea malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-cowpea malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-bambara g

The **zinc** content of the fermented and dried complementary foods ranged from 0.71-1.82 mg/g with MB_{mf} (0.71 mg/g) and (MCm)_{df} (1.82 mg/g) having the highest and lowest values respectively. On the other hand, the extruded complementary foods had zinc contents of 0.85-1.82 mg/g with samples MB_{mf} and MC_m having the highest (1.82 mg/g) and lowest (0.85 mg/g) values respectively. The **selenium** content of the complementary food blends ranged from 0.004 to 0.390 µg/g. Samples (MC_m)_{df} and MB_{mf} had the lowest (0.004 µg/g) and highest (0.390 µg/g) values respectively. The selenium content of the unfortified complementary foods ranged from 0.004-0.017 µg/g while for the fortified samples it ranged from 0.201-0.390 µg/g. The fermented samples had a range of 0.004 to 0.245 µg/g, while the extruded samples ranged from 0.010 to 0.390 µg/g. The selenium content of the maize-bambara groundnut and maize-cowpea malt complementary foods increased significantly (p < 0.05) by 59 and 44 % respectively after extrusion. The values obtained were above the recommended dietary allowance for selenium (0.20 µg/g) for infants aged 6 – 24 months.

The **vitamin A** content of the maize-bambara groundnut and maize-cowpea malt complementary foods ranged from $1.15-2.99~\mu g$ RE/g. The samples with the lowest $(1.15~\mu g$ RE/g) and highest $(2.99~\mu g$ RE/g) values were MC_m and $(MB_mf)_{df}$ respectively. The vitamin A content of the complementary food blends decreased significantly after extrusion cooking by 12% and 15% for the cowpea and bambara groundnut blends respectively.

Table 3: Micronutrient Content of Maize-Bambara groundnut malt and Maize-Cowpea malt Complementary Foods

Samples	Calcium (mg/g)	Iron (mg/g)	Zinc (mg/g)	Selenium (µg/g)	Vitamin A (μgRE/g)	Vitamin D (IU/g)	
(MB _m) _{df}	$0.66^a \pm 0.001$	$1.71^a \pm 0.021$	$0.91^a \pm 0.021$	$0.006^a \pm 0.001$	$1.66^a \pm 0.028$	$0.006^a \pm 0.001$	
$(MB_{mf})_{df}$	$4.10^{ab} \pm 0.022$	$5.40^{ab} \pm 0.028$	$1.66^a \pm 0.007$	$0.245^{ab} \pm 0.021$	$2.99^a \pm 0.014$	$0.009^b \pm 0.002$	
$(MC_m)_{df}$	$0.55^{ab} \pm 0.002$	$2.21^{ab} \pm 0.007$	$0.71^a \pm 0.014$	$0.004^{bc} \pm 0.001$	$1.45^a\pm0.035$	$0.007^{c} \pm 0.001$	
$(MC_{mf})_{df}$	$3.85^{ab} \pm 0.001$	$7.04^{ab} \pm 0.021$	$1.29^a \pm 0.014$	$0.201^{abc} \pm 0.001$	$2.77^a \pm 0.021$	$0.010^d \pm 0.001$	
MB_m	$0.7625^{ab} \pm 0.004$	$1.74^{b} \pm 0.021$	$0.96^a \pm 0.014$	$0.017^{bd} \pm 0.003$	$1.34^a \pm 0.014$	$0.003^{e} \pm 0.001$	
MB_{mf}	$4.99^{ab} \pm 0.014$	$5.87^{ab} \pm 0.021$	$1.82^{a} \pm 0.028$	$0.390^{abcd} \pm 0.014$	$2.53^a \pm 0.021$	$0.005^{\rm f} \pm 0.001$	
MC_m	$0.64^b \pm 0.002$	$2.49^{ab} \pm 0.014$	$0.85^a \pm 0.014$	$0.010^{b} \pm 0.001$	$1.15^a\pm0.028$	$0.004^g \pm 0.001$	
MC_{mf}	$4.12^a \pm 0.003$	$7.32^{ab} \pm 0.028$	$1.51^a \pm 0.007$	$0.290^{abcd} \pm 0.014$	$2.44^a \pm 0.021$	$0.008^h \pm 0.001$	
P	$3.50^{ab} \pm 0.028$	$0.08^{ab} \pm 0.007$	$0.10^a \pm 0.001$	$0.055^{abcd} \pm 0.003$	$13.00^a \pm 0.028$	$1.83^{abcdefgh}$ \pm	
						0.035	
RDA	2.50 - 5.00	0.275	0.125	0.200	5.00	4.00 - 6.00	

Key: Values are means of triplicates determinations, values carrying different superscripts in the same columns are significantly different (p<0.05). MBm: Extruded Maize-bambara groundnut malt; MBmf: Fortified and Extruded Maize-Bambara groundnut; MCm: Extruded Maize-Cowpea malt; MCmf: Fortified and Extruded Maize-Cowpea malt; (MBm)_{df}: Fermented and dried Maize-Bambara groundnut; (MBmf)_{df}: Fermented and Dried Fortified Maize-Cowpea malt; (MCm)_{df}: Fermented and Dried Fortified Maize-Cowpea malt; P: Proprietary Formula. RDA = Recommended Dietary Allowance, (Lutter and Dewey 2003.

The **vitamin D** content of the fermented and dried maize-bambara groundnut and maize-cowpea malt food blends ranged between 0.006 - $0.010 \,\mu\text{g/g}$ whereas that of the extruded complementary foods ranged from 0.003 to $0.008 \,\mu\text{g/g}$. The vitamin D content of the extruded complementary foods decreased significantly by 44 % and 20 % for bambara ground nut malt and cowpea malt blends respectively.

Phytochemical composition of the complementary foods

The phytochemical composition of the complementary foods are shown in Table 4. The **tannin** content of the complementary foods ranged from 0.272 - 0.644 mg/100g which 'is below the safe limit of 200 mg/100g. The tannin content present in bambara groundnut (0.478 mg/100g) is higher than that of cowpea (0.463 mg/100g). The tannin content of the fortified extruded foods were higher than that of the fortified fermented and dried maize-cowpea complementary foods. The **phytate** in the complementary foods ranged from 0.193 - 0.938 mg/100g and was lower than the safe limit of (250mg/100g). The **oxalate** content of the complementary food ranged from 1.114 - 1.575 mg/100g. The **phenol** content of the maize-bambara groundnut malt and maize-cowpea malt complementary foods ranged from 2240.38 – 2907.79 μ g/g. The fortified samples had higher phenolic content ranging from 2240-2907 μ g/g as against the 2300-2640 μ g/g for the unfortified samples. The **flavonoid** content of the complementary foods ranged from 167.33 – 566.57 μ g/g. The **alkaloid** content of the infant foods ranged from 0.33 – 2.47 μ g/g and were significantly (p < 0.05) higher than the value for the proprietary formula. The results show that fermentation significantly decreased the alkaloid content of the maize-bambara groundnut malt and maize-cowpea malt complementary foods by 49.80% and 82.43% respectively.

Table 4: Phytochemical Composition of the Complementary Foods

Samples	Tannins (mg/100g)	Oxalate (mg/100g)	Phytate (mg/100g)	Phenols (µg/g)	Flavonoids (µg/g)	Alkaloids (µg/g)
$(MB_{m})_{df}$	$0.302^a \pm 0.001$	$1.439^a \pm 0.002$	$0.478^a \pm 0.004$	$2581.77^{\rm a} \pm 0.18$	$537.80^{ab} \pm 0.28$	$1.25^a \pm 0.02$
$(MB_{mf})_{df}$	$0.573^a \pm 0.001$	$1.518^{a}\pm0.004$	$0.541^{ab} \pm 0.002$	$2860.93^a \pm 0.11$	$686.98^{ab} \pm 0.17$	$2.48^a \pm 0.01$
$(MC_m)_{df}$	$0.404^a \pm 0.001$	$1.495^a \pm 0.003$	$0.653^{abc} \pm 0.003$	$2640.60^a \pm 0.14$	$456.82^a \pm 0.26$	$1.11^a \pm 0.03$
$(MC_{mf})_{df}$	$0.645^a \pm 0.001$	$1.577^a \pm 0.002$	$0.937^{abc} \pm 0.002$	$2907.79^a \pm 0.09$	$566.57^{ab} \pm 0.19$	$2.03^a \pm 0.04$
MB_{m}	$0.274^a \pm 0.002$	$1.116^a \pm 0.003$	$0.195^{abc} \pm 0.002$	$2075.10^a \pm 0.07$	$216.66^b \pm 0.27$	$0.48^{ab} \pm 0.02$
$MB_{mf} \\$	$0.540^a \pm 0.003$	$1.164^a \pm 0.003$	$0.223^{abc} \pm 0.001$	$2240.38^a \pm 0.17$	$232.52^b \pm 0.34$	$0.96^{ab}\pm0.01$
MC_{m}	$0.384^a \pm 0.003$	$1.145^a \pm 0.001$	$0.359^{abcd}\pm0.002$	$2300.73^a \pm 0.25$	$167.33^a \pm 0.15$	$0.34^{ab}\pm0.01$
MC_{mf}	$0.455^a \pm 0.001$	$1.172^a \pm 0.001$	$0.566^{ad} \pm 0.066$	$2504.92^a \pm 0.12$	$181.51^a \pm 0.10$	$0.73^{ab}\pm0.03$
P	$0.467^a \pm 0.001$	$1.114^a \pm 0.002$	$0.312^{abce}\pm0.002$	$2407.43^a \pm 0.21$	$128.605^a \pm 0.29$	$0.52^a \pm 0.02$
Permissible Limit	200	500	250	-	-	-

Key: Values are means of duplicate results, values carrying different superscripts in the same columns are significantly different (p<0.05). MB_m : extruded maize-bambara groundnut malt; MB_{mf} : fortified and extruded maize-bambara groundnut; MC_m : extruded maize-cowpea malt; MC_{mf} : fortified and extruded maize-cowpea malt; $(MB_m)_{df}$: fermented and dried maize-bambara groundnut malt; $(MC_m)_{df}$: fermented and dried fortified maize-cowpea malt; $(MB_{mf})_{df}$: fermented and dried fortified maize-cowpea malt; $(PP_m)_{df}$: fermented and dried fortif

Discussion

Root Length: In bambara groundnut, root outgrowth was evident within the first 48 hours while in cowpea, root outgrowth was noticeable within the first 24 hours. The root length increased from 0.93 to 3.05 cm and 0.33 to 1.76 cm in bambara groundnut and cowpea respectively. This increase could be attributed to the secretion of gibberellins (Uvere and Uchenna, 2011) and the translocation of nutrients for growth. Attaugwu and Uvere, (2017b) reported that the increase in root length of the legumes indicated enzyme secretion and activity which led to modification of the germinating seed's endosperm.

Malting Loss and Diastatic Activity

The malting loss was 18% in bambara groundnut malt and 16.55% in cowpea malt which may be due to leaching of nutrients during steeping and degree of hydrolysis of macromolecules during germination (Uvere et al., 2010; Attaugwu and Uvere, 2017b), increased metabolic activity during germination and derooting after drying (Ayernor and Ocloo, 2007).

The diastatic activity of the bambara groundnut malt and cowpea malt increased from 44.5 - 45.20°L and 45.50 - 45.67°L after 72 hours of malting. This represents increases of 1.57% and 0.37% in the bambara groundnut malt and cowpea malt respectively. The increase is attributed to the physicochemical changes observed from the hydrolytic enzyme activities and/or the enzyme activation mechanism present in the grains (Ayernor and Ocloo, 2007). Malted bambara groundnut had higher diastatic activity with an of increase of 1.57% as compared to that of cowpea (0.37%). This is in agreement with the fact that enzymes are synthesized in proportion to the amount of available substrate present in the grain (Attaugwu and Uvere, 2017a; Uvere and Uchenna, 2011).

Proximate Composition of the Fortified Complementary Food Blends. *Moisture Content:*

The moisture content of the fermented and dried complementary foods ranged from 4.07 - 4.58 g/100g, while that of the extruded complementary foods ranged from 3.60 - 3.86 g/100g and the reference sample had a value of 3.00 g/100g. The low moisture content of the samples could result from the hydrolysis of starch and protein during malting and fermentation which led to easier loss of water during drying (Uvere et al., 2010). The further decrease in moisture content during extrusion can be attributed to the extruder barrel temperature and moisture loss as the extrudates came out of the die (Yusuf et al., 2018). The low moisture content of the complementary foods suggests a good keeping quality if appropriately stored.

Fat Content

The fat content of the complementary food ranged from 3.33-12.00 g/100g. There was a significant increase in the fat content of the complementary foods with inclusion of the fortificant mix containing the edible termite which has a fat content of 44.82g/100g (Kinyuru et al., 2013). Sample (MC_{mf})_{df} had the highest fat content as compared to MB_m . There was a decrease in fat content on extrusion cooking (value range): this a number of factors may be responsible for this: (a) complex formation between the lipids and starch and/or protein during processing which contributed to resistance of lipid extraction (Leszek, 2011) and/or (b) loss of fat due to rendering during extrusion cooking. This result agrees with the report of Yusuf et al., (2018) in which the fat content in blends of sorghum, groundnut and tigernut mix also decreased on extrusion. The reduced fat content may also indicate a reduced possibility of lipid oxidation which can affect the sensory and nutritional quality of the product.

Crude Protein Content

The crude protein content of the complementary foods ranged from 14.78 to 32.69g/100g. The fermented and dried complementary foods had a significant (p < 0.05) increase in crude protein with the inclusion of the fortificant mix, which included the edible termite which has a crude protein content of 33.51g/100g (Kinyuru et al., 2013). Other factors which resulted to the increased crude protein content are the increase in microbial biomass during fermentation and protein contributions from the 18g/100g in bambara groundnut (Ndidi et al., 2014) and 32g/100g of cowpea (Jayathilake et al., 2018). Upon extrusion, there was also a significant increase in the protein content of the complementary foods which may be attributed to removal of more moisture from the food. This result is in agreement with the report of Onwulata et al., (2001) that showed that inclusion of protein-rich materials in extruded food product improves the protein quality and utilization of the food product. The high content of lysine and leucine in bambara ground nut at 5.91 g/100g and 6.92 g/100g respectively (Kgaogelo et al., 2023), in cowpea at 6.8 g/100g and 7.7 g/100g (Affrifah et al., 2021) and termite at 6.16 g/100g and 7.76 g/100 g respectively (Inje et al., 2018) would play a possible role in boosting the immunity of the consuming infant population.

Ash Content

The ash content of the formulated complementary foods increased as a result of fortification, fermentation and extrusion cooking (2.60-5.28g/100g) and was higher than that of the proprietary formula (2.50g/100g). The increase in the fermented and dried complementary foods could indicate that pre-fermentation fortification improved the degradation of organic matter (Obizoba and Atti, 1991) which is high in termites (Fombong and Kinyuru, 2018). This could result from the availability of nutrients for microbial growth.

Crude Fiber Content:

The crude fiber in the complementary foods was low and ranged from 1.17 - 1.89 g/100g with $(MC_{mf})_{df}$ having the highest value and MB_m the lowest. The low crude fibre content of the complementary foods could be attributed to the reported low crude fibre content of 3.50g for bambara groundnut (Eltayeb et al., 2011 and 4.67g for cowpea (Mune et al., 2007). The lower crude fiber content of the complementary foods (1.17 - 1.89g) compared to the proprietary formula (4.50g) may be as a result of the use of dehulled ingredients in formulating the complementary foods. In the extruded samples, the crude fiber content

significantly decreased (p < 0.05) because larger fragments of fiber may have been sheared off during the extrusion process (Leszek, 2011).

Carbohydrate Content:

The carbohydrate content of the complementary foods ranged from 48.05 - 70.47g. There was a decrease in the carbohydrate content of the fermented and dried samples on inclusion of the fortificant mix. This may be due to the winged termite present in the fortificant (Ezeocha et al., 2020). The carbohydrate content of the extruded complementary food also increased significantly (p <0.05) due possibly to the digestion and gelatinization of the starch into dextrin and simple sugars (Wang et al., 1991) as a result of fermentation, the barrel temperature, shear pressure and shear rate (Yusuf et al., 2018).

Calcium, Iron, Zinc, Selenium, Vitamin A and Vitamin D content of the Maize-Bambara groundnut malt and Maize-Cowpea malt Complementary Foods.

The **calcium** content of the samples ranged from 0.55 - 4.99 mg/g with samples (MC_m)_{df} and MB_{mf} having the lowest (0.55 mg/g) and highest (4.99 mg/g) values. The proprietary formula contained a calcium content of 3.50 mg/g which was lower than the fermented and dried fortified maize-bambara groundnut malt (4.10 mg/g), fermented and dried fortified maize-cowpea malt (3.85 mg/g), extruded maize-bambara groundnut malt (4.99 mg/g) and extruded maize-cowpea malt (4.12 mg/g) complementary foods. There was a significant increase in the calcium content of the fortified fermented and dried complementary foods as compared to the unfortified blends. This may be as a result of hydrolysis of the anti-nutrients in the food by enzymes in the malted grains and the fermenting microorganisms, thus, making the micronutrient more bioavailable (Oladele and Oshodi, 2008). Further contributions to the increased calcium content could be from the termite (*Macrotermes bellicosus*) and hibiscus calyces (*H. sabdariffa*). The calcium content of the extruded samples also increased significantly (p < 0.05) by 16.4 - 21.7% due to increased bioavailability (Yusuf et al., 2018) and may be due to the barrel temperature and screw speed of the extruder (Gulati *et al.*, 2018). The fortified complementary foods had calcium contents within the RDA range (2.50 – 5.00 mg/g) for infants aged 6 - 24 months. This result agrees with that of Makinde et al., (2013); that reported similar increases in the calcium content on fermentation and extrusion cooking of sesame seeds and Acha.

The **iron** content of the complementary foods ranged from 1.71 to 7.32 mg/g. The fermented and dried samples had iron contents ranging from 1.71 to 7.04 mg/g while that of the extruded complementary food ranged from 1.74 to 7.32 mg/g. The fortified complementary foods had higher iron content (5.40 - 7.32 mg/g)mg/g) compared to the proprietary formula (0.08 mg/g) and were within the recommended dietary allowance for infants aged 6-24 months (0.275 mg/g). The increased iron content of the fortified complementary food blends could be traced to the inclusion of hibiscus (H. sabdariffa) calyces with iron content of 345 mg/g (Attaugwu et al., 2017). The results also showed a significant (p < 0.05) increase in the iron content (1.8 to 4.0%) of the complementary foods on extrusion cooking. This increase could have been influenced by the reduction of tannins and phytate at the extrusion temperature of 110°C making them more bioavailable. A similar report on increased iron content and bioavailability of Great Northern beans after extrusion cooking due to reduced phytate content was published by Gulati and Rose, (2018). The maizecowpea blend had a higher iron content than the maize-bambara groundnut complementary foods. This could be as a result of the effect of malting (Attaugwu and Uvere, 2017a) and the higher iron content in cowpea of 8.27 mg/100g (Affrifah et al., 2021) compared to the bambara groundnut value of 2 mg/100g (Maphosa et al., 2022). The high iron content of the complementary food implies that infants consuming it will have proper neurological development and production of new red blood cells (Cerami, 2017). Samples (MB_m)_{df} and MC_{mf} had the lowest (1.71 mg/g) and highest (7.32 mg/g) iron contents respectively.

The **zinc** content of the fermented and dried complementary foods ranged from 0.71 - 1.82 mg/g with MB_{mf} and $(MC_m)_{df}$ having the highest (0.71 mg/g) and lowest (1.82 mg/g) values. On the other hand, the extruded complementary foods had zinc content of 0.85 - 1.82 mg/g with samples MB_{mf} and MC_m having the highest (1.82 mg/g) and lowest (0.85 mg/g) values respectively. This increase may be due to the high zinc content in the hibiscus calyces of H. sabdariffa at 0.73 mg/g (Attaugwu et al., 2017), termites (Macrotermes

bellicosus) at 1.16mg/g (Kinyuru et al., 2013) and the effects of high screw speed (200rpm), and high barrel temperature (110°C) which may have degraded phytate-mineral complexes (Paridhi et al., 2020). The significant zinc content would result to the improved growth and development of reproductive organs, brain and immune system functioning of children (Wessels et al., 2020).

The **selenium** content of the complementary food blends (Table 3) ranged from 0.004 to 0.390 μ g/g and were within the recommended dietary allowance of 0.20 μ g/g for infants between 6 – 24 months.. Samples (MCm)_{df} and MBmf had the lowest and highest values respectively. The selenium content of the maize-bambara groundnut and maize-cowpea malt complementary foods increased significantly by 15 – 59% after extrusion. This means that feeding infants with these foods would contribute to improving immune system function against flu-like diseases (Malochleb, 2020) and cognitive performance.

The results also showed that the **vitamin A** content of the maize-bambara groundnut and maize-cowpea malt complementary foods ranged from $1.15-2.99~\mu g$ RE/g. The samples with the lowest $(1.15~\mu RE/g)$ and highest $(2.99~\mu RE/g)$ values were MC_m and $(MB_{mf})_{df}$ respectively. The vitamin A content of the fortified and extruded complementary food blends MB_{mf} (2.53 μg RE/g) and MC_{mf} (2.44 μg RE/g) were lower than the fermented and dried counterparts $(MB_{mf})_{df}$ (2.99 μg RE/g) and $(MC_{mf})_{df}$ (2.77 μg RE/g). These losses could be as a result of the high temperature (110°C) of extrusion (Riaz et al., 2009). Paridhi et al., (2020) reported similar impact of extrusion cooking on the vitamin A contents of cereals and legumes. The vitamin A content of the maize-bambara groundnut malt and maize-cowpea malt complementary foods were lower than that of the proprietary formula. this may be as a result of fortification of the the proprietary formula with vitamin A after production which is a common practice with commercially or factory produced formula.

The **vitamin D** content of the fermented and dried maize-bambara groundnut and maize-cowpea malt food blends ranged from 0.006 to 0.010 μ g/g whereas that of the extruded complementary foods decreased significantly and ranged from 0.003 to 0.008 μ g/g. Vitamin D is unstable to heat and trace minerals such as selenium (Raiz et al., 2009).

Phytochemical Composition of the Complementary Foods

The **tannin** content ranged from 0.272 - 0.644 mg/100g which was below the safe limit of 200 mg/100g (ref). The tannin content of the fermented and dried maize-cowpea complementary foods was higher than that of the fermented and dried bambara groundnut infant foods. This is suggestive that household processing methods such as soaking, dehulling, germination and fermentation have more reductive effects on maize-bambara groundnut malt tannins than maize-cowpea malt tannins (Hussain et al., 2011). Extrusion of the complementary foods also reduced the tannins significantly (p < 0.05) possibly resulting from thermal degradation of the molecules. This could result to improved digestion and absorption of various nutrients from foods (Popova and Mihaylova, 2019).

Phytate in the complementary foods ranged between 0.193 and 0.938 mg/100g and was lower than the safe limit of 250mg/100g (Maseta et al., 2016). The low phytate content in the formulated complementary foods could be attributed to the effects of soaking and malting (Attaugwu and Uvere, 2017a) and organic acids produced during fermentation in stimularing the production of and activation of phytases in the grains. Inclusion of the fortificant mix in the cereal-legume complementary foods increased the phytate content of the blends due to the phytic acid content (102.6 mg/kg) of the termites (Ntukuyoh et al., 2012). Extrusion cooking had a significant (p < 0.05) lowering effect on the phytate content of the complementary foods due to thermal decomposition (Kaur et al., 2013) as had also been similarly reported by Ndidi et al., (2014).

The **oxalate** content of the complementary foods was low and ranged from 1.114 - 1.575 mg/100g due possibly to soaking and fermentation (Attaugwu and Uvere, 2017a). On extrusion however, the oxalate content decreased significantly (p < 0.05) due to decomposition at the extrusion temperature (110°C). This result is in accordance with the results of Eze, et al. (2020).

The **phenolic** content of the maize-bambara groundnut malt and maize-cowpea malt complementary foods ranged from $2240.38-2907.79~\mu g/g$. On extrusion cooking, the phenolic content increased due to the high extrusion temperature and shear force which may have destroyed the ester bond between phenolics and cell walls functional groups derived from proteins and cellulose amongst others (Hu et al., 2018). The recommended dietary phenolic intake is approximately 1g per day (Shahidi and Ambigaipalan, 2015) and the formulated complementary foods had values less than this permissible limit.

The **flavonoid** content of the complementary foods ranged from $167.33 - 566.57 \,\mu\text{g/g}$. The addition of fortificants increased the flavonoid content of the fortified and dried fermented complementary. However, extrusion cooking decreased the flavonoid content of the maize-bambara groundnut malt and maize-cowpea malt complementary foods due to the high extrusion temperature of 110°C and screw speed (200 rpm which may have resulted in degradation of the flavonoids (Moussa-Ayoub et al., 2015).

The **alkaloid** content of the infant foods ranged from $0.33 - 2.47 \,\mu\text{g/g}$ and were significantly (p < 0.05) higher than the value for the proprietary formula. The results showed that steeping and fermentation significantly decreased the alkaloid content of the maize-bambara groundnut malt and maize-cowpea malt complementary foods by 49.80% and 82.43% respectively. Since alkaloids are sensitive to oxidation and are water soluble, leaching and oxidation of the alkaloids during processing may explain the decreases. On extrusion cooking, the alkaloid complexes were degraded due to the high extrusion temperature (Ruiz-Gutierrez et al., 2017). The alkaloid content of the infant foods was within the permissible level of \leq 1g per day (Okudu et al., 2017).

Conclusion

Fortifying maize-bambara groundnut malt and maize-cowpea malt complementary foods with termites and Hibiscus calyces improved the iron, zinc, selenium and calcium contents of the complementary foods. The extruded complementary food samples had higher contents of Ca, Fe, Zn, and Se compared to the fermented and dried complementary foods. Sample MB_{mf} had the highest calcium, zinc and selenium contents as compared with sample MC_{mf} which had the highest iron content of 7.32 mg/g. In comparison with the recommended dietary allowance (RDA) for infants within 6 – 24 months the processing methods were effective in releasing and concentrating the micronutrients from the fortificants with the result that they had values of 5.00 mg/g for Ca; 0.125 mg/g for zinc; 0.275 mg/g for iron and 0.2 μ g/g for selenium. Selenium have been reported to be an immune booster against flu-like viruses and at the 0.21-0.390 μ g/g content in the fortified complementary foods, they had values above the RDA (0.2 μ g/g.) for infants aged 6-24 months and could contribute to boosting the immune system of infants against flu-like diseases.

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