COMPARATIVE EVALUATION OF NUTRITIONAL QUALITY OF FERMENTED WHITE MAIZE (*Zea mays* L.) FORTIFIED WITH LIMA BEAN (*Phaseolus lunatus* L.) FLOUR

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Abstract

Ogi is a maize product which is traditionally prepared by natural fermentation, followed by wet milling, sieving and souring slurry for 2 3 days rest at room temperature. In this study, the nutritional quality ofogifrom a composite mixture of white maize (Zea mays L.) and lima bean (Phaseolus lunatus L.) flours was evaluated using standard processing techniques. Maize flour was substituted with lima bean seeds flour at ratios of 90:10, 80:20, 70:30 and 60:40 maize: lima bean; with 100% maize ogi flour as control. The results showed that moisture, ash, crude fat and crude protein contents increased progressively with increased lima bean flour substitution, reaching 89.7%, 230.5%, 257.1% andl91.6% dry weight, respectively at 60:40 ratios.Itwasfbundthatallthefortifiedproductswere good sources ofmacrominerals (Na, K, Ca, Mg and PJ *while* P *recorded increased concentration in the ogi flours with increased Hmu bean flour substitutions. Harmful heavy metals such as Pb and Cd were below detection limit of* OK *AAS. The total essential amino adds (TEAA) ranged from 18.78 to 28.96 gAOOg crude protein or from 39.77 to 40.94% of the iotal amino acid while the limiting amino adds (LAA) were Lys or Met + Cys* (TSAA) *as it appliedin the various samples. It was also found that fortified products had progressive increase in the concentration levels of total AA (TA.A), total nonessential AA (TNEAA), total essential AA (TEAA), essential alphatic AA (EAAA), essential aromatic AA* (EArAA) *and total sulphur AA (TSAA). Generally, the present study indicates that at equal to* 30% *lima bean flour substitution of the ogi mass, the quality attributes of ogi can be maintained with higher nutrient content.*

Keywords: Fc/mented maize, lima bean, supplementation, nutritional quality

INTRODUCTION

Most traditional weaning foods in developing countries are made from cereals, starchy fruits, roots and tubers. Such foods are often of poor protein quality and have high paste properties (Desikachar, 1979). Many Nigerians are now consuming less quality foods at the expense of what they need for a healthy life (Mbata *et al*, 2009). The weaning period (between 4 and 6 months after birth) is a very important phase in the life of a child. It is a period during which a variety of nonmilk foods are introduced, while the relative importance of breast milk is on the decline. It is also described as the period of transition from an exclusively milk diet to the complete range of foods taken by the adult section of the community (Jelliffe, 1973). Children in most developing countries, particularly those in lowincome classes, are weaned on cheap, readily available starchy foods. This can be attributed to several factors including poor nutritional education, decline in household incomes and the unavailability of nutrition commercial formulae (Enujiugha, 2006). This has led to the continued research on locally sourced cheap proteinrich foods and their incorporation into traditional staples. Efforts are also made to ferment the local starchy staples in order to raise their nutritional status.

In Nigeria and other parts of West Africa, cereal grains lack two essential amino acids, lysine and tryptophan (FAO/WHO/UNU, 1985; Hoshiai, 1991), thus making their protein quality poorer compared to that of animals (Chavan and Kadam, 1989). Earlier studies have documented increased lysine and tryptophan in germinated corn (Aremu, 1993), improved vitamin content in germinated sorghum and maize (Asiedu *et al.*, 1993), increased amino acid, minerals and vitamins in fermented blends of cereals and legumes (Onilude *et al.*, 1999; Enujiugha, 2006; Aremu *etal*, 2011a&b).

Attempts are currently made, in the developing world where maize, a major staple, to supplement it

\ certain unconventional legume and oil seeds contain adequate amounts of the limiting > acids. One such legume source is lima bean >*lus lunatus* L.) seeds. Although this articular legume seed is deficient in llphurcontaining amino acids, cystine and nine (which are abundant in maize), its ! and tryptophan contents are quite adequate [(Aremu *et al.,* 2010a), making it an excellent of the protein quality of maizebased ducts. With a protein content of 22.2 g/lOOg the lima bean seed has been viewed as alnutrition (PEM) experienced in i third world (Hurrel *et al,* 2003; Aremu *et al,* lOa). The defatted seed is expected to supplement ! monotonous cerealbased porridges consumed I in the west coast of Africa.

line lima seed, like some other legumes and [oilseeds, is known to contain high concentrations of flectin, trypsin, inhibitors, phytates, tannins and loxalates (Adeyemi and Soluade, 1993; Enujiugha Agbede, 2000; Aremu *et al,* 2009), but these \ tnitnutritional factors are usually removed or | greatly reduced during hydrothermal treatment, i soaking and fermentation which the seeds are [subjected to during processing (Aremu *et al.,* |2010b). However, it remains to be known how | much of the treated seed is needed by a weaningage child without negative nutritional effects. Therefore, the objective of this work was to evaluate proximate, mineral and amino acid compositions of fermented white maize *(Zea mays* L.) locally known as "ogi" fortified with lima bean *(Phaseolus lunatus* L.) flour with a view to providing preliminary information towards utilization of this legume in various food applications in Africa.

MATERIALS AND METHODS

Sample Collection and Preparation

The lima bean *(Phaseolus lunatus* L.) seeds used for the study were purchased from Owo market in Ondo State, Nigeria while the white maize grains *(Zea mays* L.) were purchased from Keffi market in Nasarawa State, Nigeria. The fermented maize (ogi) and lima bean flour were produced in the laboratory as outlined in Fig. 1. Wet milling was done using laboratory Kenwood blender (MiniProcessor Model A90LD, Thorn Emi Kenwood Small Appliance Ltd., Hampshire, UK) while milling of the flours was done using Hammer Mill, to pass through a 0.25 mm screen. The dehulled lima bean seed flour was added to maize flour at substitution levels of 10, 20, 30, and 40%, respectively; 100% fermented maize flour serve as control. The fortified flours were then packaged in moistureproof, airtight polyethylene containers and kept at 4°C prior to analyses.

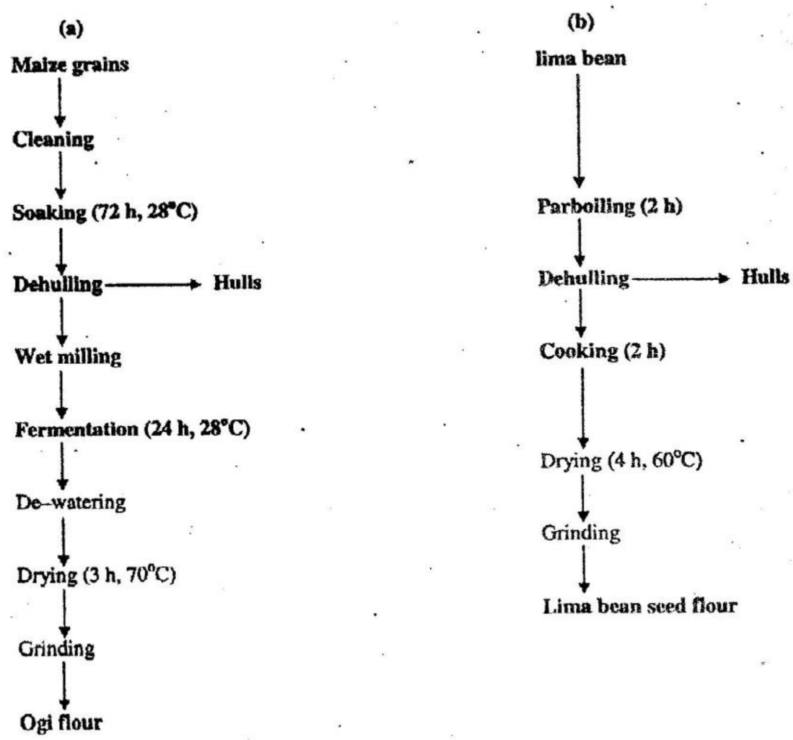

Fig. 1(ab): Processing methods adapted for the production of (a) Ogi and (b) Lima seed flours

Proximate Analysis

The moisture, ash, ether extract, crude fibre, crude protein (N x 25) and carbohydrate (by difference) were determined in accordance with AOAC (1995) methods. All proximate analyses of the sample flours were carried out in triplicate and reported in percentage. All chemicals were of Analar grade.

Mineral Analysis

The minerals were analyzed by dryashing the samples at 500°C to constant weight and dissolving the ash in volumetric flask using distilled, deionized water with a few drops of concentrated hydrochlorine acid. Sodium and potassium were determined by using a flame photometer (Model 405, Corning, UK) using Nad and KC1 to prepare the standards. All other metals were determined by atomic absorptionspectrophotometer (PerkinElmer Model 403, Norwalk, CT, USA). The minerals were reported in mg/lOOg sample.

Anuno Acid Analysis

Amino acid analysis was by Ion Exchange Chromatography (IEQ (FAO/WHO, 1991) using the Technicon Sequential Multisample (TSM) Amino Acid Analyzer (Technicon Instruments corporation, New York). The period of analysis was 76 min for each sample. The gas flow rate was 0.50 mL/min at fc>0°C with reproducibility consistent within 3%. The net height of each peak produced by the chart recorder of the TSM (each representing an amino acid) was measured and calculated. The amino acid values reported were the averages of two determinations. Norleucine was the internal standard. Try ptophan was not determined.

Estimation *cf* Isoelectric Point (pi), Quality of Dietary ProKin and Predicted Protein Efficiency Ratio (FPER)

The predicted isoelectric point was evaluated as follows (Olaoie and Akiiitayo, 2000):

pim=

Where: pirn is the isoelectric point of the mixture of amino acids, pli is the isoelectric point of the i* amino acids in the mixture, and Xi is the mass or mole fraction of the amino acids in the mixture.

The quality of dietary protein was measured by finding the ratio of available amino acids in the flour sample with need expressed as a ratio (FAO, 1970). Amino acid score (AAS) was then estimated by applying the following formula (FAO/WHO, 1991):

$$AAC = \frac{\text{»ig of a min o acid in Ig of test protein}}{\text{amino acid in Igof reference protein}} \times \frac{100}{1} \text{ mgoj}$$

The predicted protein efficiency ratio (PPER) of fortified flour was calculated from their amino acid composition based on the equation developed by Alsmeyerefo/. (1974). PPER - 0.468 + 0.454 (Leu) 0.105 (Tyr)

Calculation and Statistical Analysis

Calcium/phosphorus (Ca/P), sodium/potassium (Nfa/K) and Leu/Ile ratios were calculated for the samples (Nieman *et al*, 1992; Olaofe *et al*, 2008; Adeyeye, 2010). The fatty acid values were obtained by multiplying crude fat value of each sample with a factor of 0.8 (i.e. crude fat x 0.8 • corresponding to fatty acids value) (Paul & Southgate, 1978). The energy values were calculated by adding up the carbohydrate x 17 kj, crude protein x 17 kj and crude fat x 37 kj for each of the samples (Kilgore, 1987). Errors of three determinations were computed as standard deviation for the proximate and mineral composition.

RESULTS AND DISCUSSION

The proximate composition of fermented maize fortified with lima bean is shown in. Table 1. Increased lima bean seeds flour substitution gave progressively higher moisture, ash, crude protein and oil contents of the product with lower crude fibre and carbohydrate contents. It has been reported (Enujiugha, 2006; Aremu, 1993) that the traditional method of ogi processing is usually accompanied by severe nutrient losses which aggravates the poor nutritional quality of normal dent corn. The progressive increase observed in moisture content of the fortification product could mean a decrease in predisposition to shelf spoilage {Aremu *ei al,* 2006). There was progressive decrease in the calculated metabolizable energy values of lima bean seedsiibstituted fermented maize flours and the difference in each product was significant (p > 0.05). Lima bean is known to have containing energy concentration favourably compare to cereal (Aremu *et al.*, 2010a). Despite the effect of fortification processes the coefficient of variation (CV%) levels were relatively close with hot spot at 57.75 in energy whereas others ranged from 6.93 in carbohydrate to 39.29 in crude fat (Table 1).

Differences in the mean proximate composition between fermented maize and different fortification products are presented in Table 2. All the proximate composition parameters were progressively increased with different fortification products except crude fibre, carbohydrate and calculated metabolizable energy contents. The most affected by lima bean seedsubstituted fermented maize flours were crude protein and fat contents which were increased between 67.3-91.6% (CV%, 32.39) and 62.5 - 257.1% (CV%, 52.97), respectively. This result is similar to the one reported by Aremu *et al.* (2011a) on kersting's groundnut maize ogi fortified product. Range of reduction of crude fibre, carbohydrate and energy were 11.2-51.4%, 6.5-19.4% and 6.4-10.5%, respectively. The CV% was variously varied with a range of 22.68-52.97 (Table 2). .

The mineral composition of the weaning diets is presented in Table 3. The macrommerals such as Na, K, Ca and Mg recorded reduced concentrations in the fermented maize with increased lima bean flour substitutions while P increased in concentration with increased substitution ratios. However,, tins report is contrary to the report of Aremu *et al* (2011b) who observed increased in concentrations of Mg and Ca for fermented white maize blended with scarlet runner bean. This reason could be due to different processing methods (Enujiugha, 2006; Aremu *et al*, 2011a). Magnesium is an important element in connection with circulatory diseases such as ischemic heart disease and calcium metabolism in bone (Fleck, 1976). Calcium in conjunction with phosphorus, magnesium, vitamins A, C and D, chlorine and protein are all involved in bone formation (Shills, 1973). Calcium is also important in blood clotting, muscle contraction and in certain enzymes in metabolic processes (Shills, 1973). Phosphorus assists calcium in many body reactions although it also has independent function. Modern diets which are rich in animal proteins and phosphorus may promote the loss of calcium in the urine (Shills and Young, 1992). All the microminerals analyzed for (Fe, Cu, Zn, Mn, As & Al) recorded a decrease in concentrations. It is interesting to note that harmful minerals such as Cd and Pb were not at detectable range of AAS for any of the samples (Table 3). Cadmium and lead even at low concentration are known to be toxic and have no known function in biochemical process. Lead can impair the nervous system and affect foetus, infants and children resulting in lowering of intelligent quotient (IQ) even at its lowest dose (UN, 1998). Arsenic was detected in 70:30 ratio (0.02 mg/lOOg) and 60:40 ratio (0.01 mg/lOOg). Arsenic has been implicated in lung cancer, especially when the arsenic compound inhaled is of low solubility. It has also been found to have an effect on the liver by causing a disease termed cirrhosis and a rare form of liver cancer called haemongioendotherlioma (Hutton, 1987). The CV% levels of all the minerals were also relatively close with the highest found in Mn (85.15) and the least was Na (10.41) (TableS).

Differences in die mean mineral composition between fermented maize and different lima bean substitutions showed that CV% ranged from 16.25 in Zn to 83.15 in Cu (Table 4). Overall, it could be concluded that the substitution of fermented maize with lima bean seeds in the production of ogi, a weaning diet, yield products that were rich in mineral content despite the loss of nutrient during processing. The lima bean seed is known to contain appreciable amounts of important minerals (Aremu *et al,* 2010a; Oshodi *et al,* 1998) and this is expected to reflect in any legumebased or supplemented food product. Food is considered good if Ca/P ratio is above one and poor if the ratio is less than 0.5 while Ca/P ratio above two helps to increase the absorption of calcium in the small intestine. The results of Ca/P ratios in fermented maize sample and fortified products (Table 3) were not only good but also gave an indication that they

Table 1: Mean proximate composition (%) of fermented maize fortified with lima bean flour

Parameter	100% Fermented Maize I	Fermented Maize : Lima bean						
		90:10 II	80:20 III	70:30 IV	60:40 V	Mean	SD	CV (%)
Moisture	5.35(0.25)"	7.80(0.19)	8.45(0.12)	9.60(0.05)	10.15(0.13)	8.27	1.68	20.29*
Ash	0.95(0.14)	1.78(0.10)	2,11(0.04)	2.86(0.35)	3.14(0,09)	2.17	0.78	36.06
Crude fat	0.56(0.02)	0.19(0.08)	1.64(0.19)	1.84(0.15)	2.00(0.25)	1.39	0.55	39.29
Crude protein	6.21(0.33)	10.41(005)	16,91(0.44)	17.17(0.23)	18.11(0.15)	13.76	4.66	33.89
Crude fibre	2.14(0.25)	1.90(0.15)	1.54(0.34)	1.16(0.10)	1.04(0.08)	1.56	0.42	26.94
Carbohydrate"	77.48(1.01)	72.44<1.4S)	70,1<KJL23)	69.31(1.23)	62.44(2.35)	10.36	4.88	6.93
Fatty acid?	0.45(0.20)	0.73(0.12)	1.31(0.25)	1.47(0.40)	1.60(0.55)	1.11	0.44	39.26
Energy"	1579.44 (6.35)	1514.26 (5.87)	1478.67 . (7.32)	1436.23 (4.34)	1417.44 (6.56)	1485.21	3.89	57.75

"Number in parentheses are standard deviations of triplicate determinations; "Carbohydrate percent calculated as the (100 - total of other components); 'Calculated fatty acids (0.8 x crude fat); ^Calculated raetabolizable energy kJ/lOOg (protein x 17 •+• fat x 37 + carbohydrate x 1.7)

Table 2s Differences » the meats proximate composition between fermented maize with •< different fortified products ;;

Parameter	i-n	• i-ni	i-IV	;' I-V-	Mean	SD	CV (%)
Moisture	-2.45(-4S.8%)	-3.1Q(-S8,0%)	-4.25(-79.4%)	-4.80(89.7%)	3.65	0.93	22.68
Ash	-Q.83(-87.4%)	-U6(-122.1%)	-1.91(-20l.l%)	-2.19(-230.5%)	1.52	0.55	36,13
Crude fat	-0.3S(-62.52%)	-l.08(-192.9%)	-1.28(-228.6%)	-1.44(-257.5%)	1.04	0.55	52.97
Crude protein	-4.20HS7.3%)	-1Q.70H72.3)	-10.96(-176.5%)	-11.90(-1SH.6%)	9.44	3.06	32.39
Crude fibre	0.24(11.2%)	0.60(28.0%)	0.98 (45.8%)	1.10(51.4%)	0.73	0.34	46JZ7
Carbohydrate	5.04 (6.5%)	7.34(9.5%)	8.17(10.5%)	15.04(19.4%)	8.90	3.73	41.88
Fatty acid	-G.28(~62.2%)	-0.86(-191.1%)	-1.02(-226.7%)	-1.15(-255.6%)	0.83	0.33	40.05
Energy(kJ/100g)	165.18(10.5%)	100.77(6.4%)	143-21(9.1%)	162.00(10.3%)	142.79	25.67	17,98

1 ss 100% fermented maize; II = 90FM: 10LB; III a 80FM: 20LB; IV = 70JFM: 30LB; V = 60FM: 40LB; FM•«= fermented maize; LB = lima bean; SD =s standard deviation; CY s coefficient of variation • .

Table 3: Mean mineral composition (mg/lOOg) of fermented make fortified **with Urns bean flour**

Mineral	I	II	IH	IV	• V	Mean	SD	CV(%)
Na	83.10(0.03)	81.01(0.02)	68.10(1.00)	68.25(0.50)	.64.30(1.55)	72.95	7.60	10.40
K	331.50(1.20)	304,50(0.02)	301.10(1.30)	241.7.0(1.20)	240.0UJ.OO)	284.66	36.70	12^93
CU	246.50(1.10)	214.20(1.50)	182.25(1,45)	I63,20(,t.20)	131.20(0.50)	187.47	41.78	22.28
Mg	221.24(2.10)	115.50(2.20)	135.10(1.60)	133.25(0.40)	125.10(0.60)	146.04	3S.2	26.18
P	15.20(0.50)	17.30(0.20)	25.24(2.50)	56.35(1.20)	64.25(1.30)	35.67	20.54	57.59
Fe	11.10(0.50)	10J7K0.30)	9.20(0.25)	6.31(0.24)	9.45(0.20)	9.02	1.72	19.04
Cu	3.10(0.10)	3.02(0.10)	2.40(0.01)	3.01(0.03)	2.11(0.01)	2.73	0.40	14.59
Zn	17.21(2.50)	9.24(0.70)	8.50(1.50)	6.40(0.80)	5.23(0.25)	9.32	4.20	45.06
Mn	35.20(2.50)	22.14(1.10)	10.15(2.50)	2.70(0.30)	3.01(0.50)	14.64	12.46	85.15
As	ND	ND	ND ,	0..02 (6.01)	0.01(0.01)	nd	nd	nd
Al	1.32(0.10)	. 0.34(0.21)	0.30(1.00)	U8(0.10)	0.18(0.02)	0.66	0.48	7354
n	ND	ND	ND. :	;' . ND	*m*	nd	nd	Bd-:.:.:
CW '	16.23	12.38	7.22:	2.90	2.04	8.15	5.46	66.97
Naflt	0.25.	0.27	0.23	; 0.28:	0.27	0.26	0.02	: -TiBS: ;::-;|

Numbers in parentheses are standard deviations of Uiplicate determinations ; :|
l«aOO%FM;n*90FM: lOOl;ffl = 80FMr20LB;IV^70FM:30LB; V=s60FM:40LB; ;
FM « fermented maize; LB = lima bean; SD s standard deviation; CV s coefficient of variation; i |
ND a not detected; nd = not determined

Table 4: Differences in the mean tniiteral composition between **fermented matee and different fortified producta**

Mineral	I-II	*I -III*	i-re •	I-V	Mean	SD	cy
Na	2.09 (2.5%)	15.00(18.1%)	14.85 (17.9%)	18.8 (22.6%)	12.89	5.98	46,39
K	27.00(8.1%)	30.40(9.2%)	90.30(27.2%)	91.49(27.6%)	59.80	31.12	52.05
Ca	32.30(13.1%)	64.25 (26.1%)	83.3 (33,8%)	15.30 (46.7%)	73.79	30.25	40.99
Mg	105-75(47-8*)	86.15 (38.9%)	88.00 (39.8%)	96.15 (43.5%)	94.01	7.75	8^4
P	-ZMK-13.8%)	-10.04(-&U%)	-4U5(~?.7ft?%)	-49.05(-322.7%)	25.59	19.92	77.84
Fe	0.39(3.5%)	1.90(17.1%)	4.79 (43.2%)	1.65 (14.9%)	2,18	1.61	73,88
Cu	0.08(2.(S%>	0.70(22.6%)	0.09 (2.9%)	0.99(31.9%)	0.47	0.39	83.15
Zn	7.97(46.3%)	8.71 (0.5%)	JO.B1 (62.8%)	11.98(69.6%)	9.87	1.60	16.25
Mn	13.06 (0.4%)	25.05 (71.2%)	32.50 (92.3%)	32.19(91.5%)	25.7	7.88	30.67
As	nd	nd	nrt	nd	nd	nd	nd
Al	0.98 (74.2%)	1.02(77.3%)	0.14(0.1%)	1J,4(0.9%)	0.82	0.40	48.41
Cd	nd	nd	nd	nd	nd	nd	nd
Pb	nd	nd	nd	nd	nd	nd	nd
Ca/P	3 85 (23.7%)	9.01 (55.5%)	13.33 (82,1%)	14.19 (87.4%)	10.10	4.13	40.85
Nfl/K	-0,a(-tQ*)	fl 05 (8,0%)	4).03 *(_-4U%}*	-J0.02 (-8.0%)	0.02	0.00	0.00r

I - KWtemrf; II = 90FM : 101B; III« 80FM : 20IJ»; IV ^ 70IM : 30t-B; V = 60PM: 40LBV FM •
fcrttifeftted fflaiZS! **LB'«** Uma tean; SO =s standard dcvifltion: CV « coeflicient of variation;
nil r: inn determined

Table 5: Amino acid profiles of fermented maize with lima bean flour (g/100g crude protein)

Amino acid	I	II	III	IV	V	Mean	SD	CV(%)
Lysine (Lys)[y]	2.0	2.3	2.8	2.7	4.3	2.82	0.79	28.14
Histidine (His)[y]	1.8	2.0	2.2	2.2	2.4	2.12	0.21	9.91
Arginine (Arg)[y]	3.0	2.5	3.7	3.8	5.2	3.64	0.91	25.09
Aspartic acid (Asp)	6.4	6.8	7.0	6.8	8.9	7.00	1.02	14.60
Threonine (Thr)[y]	2.1	2.2	2.3	2.2	2.6	2.28	0.17	7.55
Senine (Ser)	2.0	2.4	3.1	2.3	3.7	2.70	0.62	22.83
Glutamic acid (Glu)	8.6	9.1	9.6	9.4	13.3	100	1.68	16.84
Praline (Pro)	1.8	2.0	2.4	1.9	3.1	2.24	0.48	21.24
Glycine (Gly)	2.0	2.1	2.1	2.2	2.5	2.18	0.17	7.89
Alanine (Ala)	1.9	2.0	2.4	2.7	3.9	2.58	0.72	27.89
Cystine (Cys)	0.7	0.7	0.8	0.9	1.1	0.84	0.15	17.82
Valine (Val)[y]	2.7	3.0	3.3	3.0	4.2	3.24	0.52	15.93
Methionine (Met)[y]	0.7	0.9	1.0	0.9	1.3	0.96	0.20	20.41
Isoleucine (Ile)[y]	2.2	2.5	3.0	2.9	3.7	2.86	0.51	17.77
Leucine (Leu)[y]	3.9	4.1	5.1	4.7	6.5	4.86	0.92	19.02
Tyrosine (Tyr)	1.6	1.8	2.1	1.9	2.2	1.92	0.21	11.12
Phenylalanine (Phe)[y]	2.8	3.2	3.4	3.3	4.1	3.36	0.42	12.57

[y]Essential amino acids; I = 100%FM; II = 90FM : 10LB; III = 80FM : 20LB; IV = 70FM : 30LB;
V = 60FM : 40LB; FM = fermented maize; LB = lima bean; SD = standard deviation;
CV = coefficient of variation;

Table 6: Differences in the mean amino acid composition between fermented maize and different fortified products

Amino acid	II	III	IV	V	Mean	SD	CV(%)
Lys[y]	−0.3(−15.0%)	−0.8(−40.0%)	−0.7(−35.0%)	−2.3(−115.0%)	1.03	0.76	73.74
His[y]	−0.20(−11.1%)	−0.4(−22.2%)	−0.4(−22.2%)	−0.6(−33.3%)	0.4	0.14	35.36
Arg[y]	−0.50(−16.7%)	−.07(−23.3%)	−0.8(−26.7%)	−2.2(−73.3%)	1.48	0.79	53.94
Asp	−0.4(−5.9%)	−0.6(−9.4%)	−0.4(−5.9%)	−2.5(−39.1%)	1.00	0.87	86.89
Thr[y]	−0.10(−4.8%)	−0.2(−9.5%)	−0.1(−4.8%)	−0.5(−23.8%)	0.23	0.16	71.31
Ser	−0.40(−20.0%)	−1.1(−55.0%)	−1.3(−56.5%)	−1.7(−85.0%)	1.13	0.47	41.69
Glu	−0.50(−5.8%)	−1.0(−11.6%)	−0.8(−9.3%)	−4.7(−54.7%)	1.75	1.71	97.85
Pro	−0.20(−11.1%)	−0.6(−33.3%)	−0.1(−5.6%)	−1.3(−41.9%)	0.55	0.47	85.76
Gly	−0.10(−5.0%)	−0.1(−5.0%)	−0.2(−10.0%)	−0.5(−25.0%)	0.23	0.16	71.31
Ala	−0.10(−5.8%)	−0.5(−26.3%)	−0.8(−42.1%)	−2.0(−195.0%)	0.85	0.71	83.40
Cys	0.00(0.0%)	−0.1(−14.3%)	−0.2(−28.6%)	−0.4(−57.1%)	0.18	0.15	82.21
Val[y]	−0.30(−11.1%)	−0.6(−22.2%)	−0.3(−11.1%)	−1.5(55.6%)	0.93	0.55	59.57
Met[y]	−0.20(−28.6%)	−0.3(−42.9%)	−0.2(−28.6%)	−0.6(−85.7%)	0.33	0.16	49.70
Ile[y]	−0.30(−13.6%)	−0.8(−36.4%)	−0.7(−31.8%)	−1.5(−68.2%)	0.83	0.43	52.09
Leu[y]	−0.20(−5.1%)	−1.2(−30.8%)	−0.8(−20.5%)	−2.6(−66.7%)	1.2	0.88	73.60
Tyr	−0.20(−12.5%)	−0.5(−31.3%)	−0.3(−18.8%)	−0.6(−37.5%)	0.4	0.16	39.53
Phe[y]	−0.40(−14.3%)	−0.6(−21.4%)	−0.5(−17.9%)	−1.3(−46.4%)	0.7	0.35	50.51

[y]Essential amino acids; I = 100%FM; II = 90FM : 10LB; III = 80FM : 20LB; IV = 70FM : 30LB;
V = 60FM : 40LB; FM = fermented maize; LB = lima bean; SD = standard deviation;
CV = coefficient of variation;

Table 7: Concentrations of essential, non–essential, acidic, basic, neutral, sulphur, aromatic, etc. (g/100g crude protein) of fermented maize fortified with lima bean flour

Parameter	I	II	III	IV	V	Mean	SD	CV (%)
Total amino acid (TAA)	46.12	49.80	56.18	53.66	72.82	55.72	9.21	16.53
Total non–essential amino acid (TNEAA)	27.34	29.56	33.18	31.88	43.86	33.16	5.71	17.22
TNEAA (%)	59.28	59.36	59.06	63.14	60.23	60.21	1.52	2.52
Total essential amino acid (TEAA)								
With histidine	18.78	20.24	23.00	21.78	28.96	22.55	3.51	15.54
Without histidine	16.98	18.22	20.82	19.60	26.56	20.44	3.32	16.24
TEAA (%)								
With His	40.72	40.64	40.94	40.59	39.77	40.53	0.40	0.99
Without His	36.82	36.59	37.06	36.53	36.47	36.69	0.22	0.59
Essential alphatic amino acid (EAAA)	16.00	17.03	19.63	18.49	24.91	19.21	3.11	16.17
Essential aromatic amino acid (EArAA)	2.78	3.21	3.37	3.29	4.05	3.34	0.41	12.28
Total neutral amino acid (TNAA)	22.85	25.32	29.07	27.08	36.30	28.12	4.57	13.21
TNAA (%)	49.54	50.84	51.74	50.47	49.85	50.49	0.77	1.53
Total acid amino acid (TAAA)	14.99	14.98	16.63	16.18	22.23	17.00	2.69	15.84
TAAA (%)	32.50	30.08	29.60	30.15	30.53	30.57	1.00	3.30
Total basic amino acid (TBAA)	6.82	7.88	8.73	8.65	11.91	8.80	1.70	19.32
TBAA (%)	14.79	15.82	15.54	16.12	15.36	15.53	1.00	6.47
Total sulphur amino acid (TSAA)	1.46	1.62	1.75	1.75	2.38	1.79	0.31	17.47
Cystine in TSAA (%)	3.17	3.25	3.11	3.26	3.27	3.21	0.06	1.93
(P-PER)[a]	1.13	1.20	1.63	1.47	2.25	1.54	0.40	25.99
(pI)[b]	3.20	3.28	3.48	3.67	3.85	3.50	0.24	6.88
Leu/Ile (ratio)	1.77	1.64	1.70	1.62	1.76	1.70	0.06	3.30
Leu/Ile (diff)	1.70	1.60	2.10	1.80	2.80	2.00	0.43	21.68
% Leu – Ile (diff)	43.6	39.0	41.2	38.3	43.1	41.04	2.12	5.17

[a](P–PER) = calculated predicted protein efficiency ratio; [b](pI) = calculated isoelectric point;
I = 100%FM; II = 90FM : 10LB; III = 80FM : 20LB; IV = 70FM : 30LB;
V = 60FM : 40LB; FM = fermented maize; LB = lima bean; SD = standard deviation;
CV = coefficient of variation

Table 8: Amino acid scores of fermented maize fortified with lima bean flour

EAA	PAAESP[a] (g/100g protein)	I		II		III		IV		V	
		EAAC	EAAS	EAAC	EAAS	EAAC	EAAS	EAAC	EAAS	EAAC	EAAS
Ile	4.0	2.16	0.54	2.54	0.64	3.01	0.75	2.89	0.72	3.65	0.91
Leu	7.0	3.85	0.55	4.10	0.59	5.05	0.72	4.67	0.67	6.50	0.93
Lys	5.5	2.00	0.36	2.32	0.42	2.84	0.52	2.67	0.49	4.32	0.79
Met + Cys (TSAA)	3.5	1.46	0.42	1.62	0.46	1.75	0.50	1.75	0.50	2.38	0.68
Phe + Tyr	6.0	4.37	0.73	4.96	0.83	5.43	0.91	5.20	0.87	6.27	1.05
Thr	4.0	2.07	0.51	2.15	0.54	2.29	0.57	2.24	0.56	2.62	0.66
Try	1.0	nd	na	nd	na	nd	na	nd	na	nd	na
Val	5.0	2.66	0.53	3.01	0.60	3.30	0.66	2.95	0.59	4.17	0.83
Total	36.0	18.57	3.64	20.70	4.08	23.67	4.63	22.37	4.4	29.91	5.85

EAA = essential amino acid; PAAESP[a] = provisional amino acid (egg) scoring pattern;
EAAC = essential amino acid composition (see Table 5); EAAS = essential amino acid score;
nd = not determined; na = not available; I = 100%FM; II = 90FM : 10LB; III = 80FM : 20LB;
IV = 70FM : 30LB; V = 60FM : 40LB; FM = fermented maize; LB = lima bean;

would help to increase the absorption of calcium in the small intestine. Table 3 also shows Na/K ratios of 100% fermented maize and fortified products. Both Na and K are required to maintain osmotic balance of body fluids, the pH of the body, to regulate muscle and nerve irritability, control glucose absorption and enhance normal retention of protein during growth (NRC, 1989). The ratio of sodium to potassium in the body is of great concern for the prevention of high blood pressure; a Na/K ratio less than 1 is recommended (Nieman *et d.,* 1992). The value of Na/K in the present study ranged between 0,23 to 0.28 (Table 3). This suggests that the fermented maize and fortified products may not promote high blood pressure if consumed (OlaofeefaZ.,2008).

Amino acid profile is presented in Table 5. Leucine was the most concentrated (3.9 to 6.5 g/lOOg crude protein) essential amino acid in all the samples (both control and fortified samples) while glutamic acid was the most concentrated amino acid (8.6 13.3 g/lOOg crude protein, cp) as expected in legumes (Aremu *et ai,* 2007, 2008; Olaofe *et al,* 2008). Tryptophan concentrations could not be determined. Increased lima bean substitutions gave progressive higher concentrations of all the essential amino acids (AA). Levels of CV% ranged from 7.55 in Tlir to 28.14 in Lys. Differences in the mean AA composition between fermented maize (100%) and fortified products revealed that the most affected substitution ratios in essential AA were Arg, Met and Lys which were increased between 16.773.3% (CV%, 53.94),' 28.6-85.7% (CV%, 49.70) and 15.0-115.0% (CV%, 73.74), respectively. Highlights of the increases recorded in other EAAs were (in %): His (11.1 - 33.3), Thr (4.8 23.8), Val (11,1 - 55.6), He (13.6 - 68.2), Leu (5.1- 66.7) and Phe (14,3 - 46,4), The CV% was variously varied with* range of 39.53 inTyr to97.85 inGlu (Table6).

The concentrations of total AA (TAA), total noncssential AA (TENAA), total essential AA (TEAA) with His, essential alphatic AA (EAAA), essential aromatic AA (EArAA) and total sulphur AA (TSAA) of 100% fermented maize were (in g/lOOg cp): 46.12,27.34,18.78,16.00,2.78 and 1.46, respectively (Table 7). Fortified samples had' progressive increase in the concentration levels of TAA, TNEAA, TEAA, EAAA, EArAA and TSAA. The contents of TEAA of 18.78, 20.24, 23.00, 21.78, 28.96 g/lOOg cp without tryptophan were close to the value for egg reference protein (56.6 g/lOOg cp) (Paul *ft al,*1976); for soya bean it is 44.4 g/lOOg cp (Adeyeye, 2010). The TAA in the present study (4612 - 72.82 g/100g cp) are close to TAA in the bambara groundnut (70.8 g/lOOg cup) (Aremu *tt al.,* 2006a) and dehulled African yam beans (703-91.8 g/lOOg cp) (Adeyeye, 1997). The contents of TSAA (1.46*2.38 g/lOOg cp) are lower than the *5i* g/lOOg cp recommendedforinfants (FAO/WHO/UNU, 1985). The EArAA range suggested for ideal infant protein (6.8 -11.8 g/lOOg cp) FAO/WHO/UNU, 1985) has current values close to the minimum (i.e., 2.78-4.05 g/lOOg cp,l The EArAA are precursors of epinephrine and thyroxin (Robinson, 1987). The percentage ratios of TEAA to TAA in the samples ranged from 39.77 to 40.94% which are above the 39% considered to be adequate for ideal protein food for infants, 26% for children and 11% for adults (FAO/WHO/UNU, 1985). The TEAA/TAA percentage contents are close to that of egg (50%) (FAO/WHO, 1991) and strongly comparable to 43.6% reported for pigeon pea flour (Oshodi *et al.,* 1993), 43.8- 44.4% reported for beach pea protein isolate (Chavan *et al,* 2001) and 40.6% reported for cashew nut (Aremu *et*«!., 2006b). The PPER values (Table 7) are comparable to pigeon pea (1.82) and cowpea (1.21) (Oyarekua and Eleyinmi, 2004) and millet ogi (1.62) (Fennema, 1985). The predicted protein efficiency ratio (PPER) is one of the quality parameters used for protein evaluation (FAO/WHO, 1991). The calculated isoelectric point (pi) ranged from 3.20 - 3.85. This is useful in predicting the pi for protein in order to -, enhance a quick precipitation of protein isolate from biological samples (Olaofe and Akintayo, 2000).

The presence of Disomers also reduces the! digestibility of the protein, because peptide bonds j involving D residues are less easily hydrolyzed h'<*vitro* than those containing only L residue,! Moreover, certain D amino acids exert a toxfci action, in proportion to the amount absorbed through the intestinal wall (Fennema, 1985). This elucidates a word of caution on the excessive; consumption of fermented maize or its fortified; products. A common feature of sorghum and maize; is that the proteins of these grains contain ii relatively high proportion of leucine. It therefore suggested that an amino acid imbalance from excess leucine might be a factor in the' development of pellagra (FAO, 1995). Clinical, biochemical and pathological observations in experiments conducted in humans and laboratory' animals showed that high leucine in the diet impairs the metabolism of tryptophan and niacm and is responsible for niacin deficiency in sorghun eaters (Ghafoorunissa and Navasinga Rao, 1973V High leucin is also a factor contributing to the, pellagragenic properties of maize (Belavady and

Gopalan, 1969). These studies suggested that the leucine/isoleucine balance is more important than dietary excess of leucine alone in regulating the metabolism of tryptophan and niacin and hence the disease process. The present Leu/He ratios were low in values (1.62-1.77). The highest variability was PPER (CV%> 25.99) while the least was % TEAA (without His) (CV%, 0.59) (Table 7).

The EAA scores (EAAS) of the samples based on the provisional amino acid scoring pattern (FAO/WHO, 1991) are shown inTable 8. The EAAS greater than 1.0 was Phe+Tyr for 60:40 substitution ratio sample. This shows that supplementation may be required in He, Leu, Lys, Met + Cys (TSAA), Thr and Val for all the fortified products. However, EAAS based on FAO/WHO/UNU (1985) Standards for the infante with a required value of 1.9 g/100g cp for His, showed that all the fortified products had scores of His more than 1.0 (Table 5). Histidme is a semiessential AA particularly useful for children growth. It is the precursor of His present in small quantities in cells. When allergens enter the tissues, it is liberated in larger quantities and is responsible for nettle rash (Bingham, 1977). The limiting amino acid (LAA) for 100% fermented maize (control), 90:10,80:20, and 70:30 substitution ratios was Lys while that of 60:40 ratio was Met + Cys (TSAA) (Table 8). This agrees with the report of FAO/WHO/UNU (1985) that the EAAS most often acting as a limiting capacity are Met (and Cys), Lys, Thr and Try. Try was not determined in this report.

CONCLUSIONS

This work has presented the nutritional composition of maize ogi from a composite mixture of fermented white maize *(Zea mays* L.) and lima bean *(Phaseolus lunatus)* seeds flour. The study has revQaled that increased lima bean flour aubetitution gave pi-6gwsaive higher moisture, ash, crude fat and qrude protein contents Of the fortified products with .lower crude fibre and carbohydrate contents. It was found that the samples were good sources of essential mincrate and of high quality protein with adequate essential amino acids expected for the infanta. However,.the quality attributes of maize ogi can h*. adequately maintained at 70:30 and 60:40 lima b«an seeds flrttir substitution with maize ogi mass ratios.

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