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Assessing The Amino Acid Profile and Nutritional Properties of Bread Produced from Wheat, Ache and Date Palm Fruit Flour Blend

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Abstract

This study aimed at assessing the amino acid profile and nutritional properties of bread produced from wheat, acha and date fruit flour blend using standard methods. The flour was formulated at different ratios of wheat: acha: date 80%:10%:10% (sample B); 70%:20%:10% (sample C) and 60%:30%:10% (sample D), with 100% wheat serving as control (Sample A) to produce the bread. Sample D (60:30:10) had the highest value of about thirteen (13) amino acids including essential amino acids. The minerals (iron, potassium, phosphorus, zinc, calcium) and vitamins (B1, B2, B6, B12 and C) all increased with corresponding increase in the level of substitution of wheat flour with acha flour and date fruit flour. There was significant difference between the control and the bread made from the blended flour samples. This study therefore deduced that healthy, nutritious and acceptable bread can be produced with up to 40% substitution of wheat flour with acha flour and date palm fruit flour. Sample D was deduced to be higher in amino acid including the sulphur amino acids (methionine and cysteine) which makes the bread sample less susceptible to denaturation, and also minerals and vitamins which are needed to reduce cases of malnutrition fight off diseases thus sample D should be recommended for baking.

Keywords: Amino acid profile; assessing; bread; flour blends; nutritional

Introduction

Bread is one of the popular daily staple foods in many countries, with refined wheat flour commonly used in most white bread formulations (Issaoui et al., 2021). Bread is a food product that is universally accepted as a very convenient form of food that has desirability of all population, rich and poor, rural and urban. Bread quality mainly depends on four parameters, that is, texture, moisture content, bread surface color, and structure (volume, shape, and size) of the bread (Panirani et al., 2023).

Flour blends are basically called composite flour which is known as mixed flours as the include starches and other ingredients that replace wheat partially and totally for the production of bakery and pastry products (Mitaigiri et al., 2021). In 1964, the FAO (Food and Agriculture Organization of the United Nations) introduced the Composite Flour Programme that aimed at the development of bakery products from locally available materials (Jisha et al., 2008). The FAO stated that the use of composite flour for fabrication of various food product would be efficiently advantageous if the significance of wheat flour could be decreased or even removed and demand for production of bread and pastry product could be met by the routine of domestically grown products as replacement of wheat (Mitaigiri et al., 2021).

Wheat (*Triticum aestivum* L) is a staple food used to make flour for leavened, flat, and steamed breads, biscuits, cookies, cake, breakfast cereal, pasta, noodles, fermented alcoholic beverages (beer) and biofuel sharma (Dubcousky and Dvorack 2007). Dubcousky and Dvorack (2007) reported further that the gluten protein fraction in wheat confers the visco-elastic properties that allowed the dough to be processed into above mentioned food products (Villarino et al., 2014; Barak et al., 2013). Shewry (2009) also explained that high content of starch (60- 70%) in wheat whole grain and relatively low protein content (8- 15%) made the crop still important source of calories and protein for human and livestock nutrition. Wheat milling contributes to increased losses of health-promoting phenolics, dietary fibers, vitamins, and minerals (Bala et al., 2023).

Acha (*Digitaria exilis*), also known as "fonio" though potentially rich in nutrients, according to Vietnameyer et. al. (1996), has been so neglected that it is called the lost crop of Africa, having received but a fraction of the attention accorded to sorghum, pearl millet, and maize (Chinwe et al., 2015). Acha does not contain any glutelin or gladin proteins which are the constituents of gluten, making it suitable for people with gluten intolerance (Ayo et al., 2007; Ayo and Andrew, 2016). The starch ration is total starch (43.6%), resistant starch (2.1%) and digestible starch (41.4%) (Jideani & Jideani, 2011) Acha is an important source of antioxidants phenolics, dietary fibre, cholesterol lowering waxes and capable of helping diabetic (Jideani & Jideani, 2011) since it has relatively low free sugar and low glycemic index (Cruz, 2004; (Jideani & Jideani, 2011, Jillian, 2020). Acha contains higher amount of Sulphur amino acids (methionine and cysteine) and is less susceptible to denaturation (Jideani & Jideani, 2011). Denaturation can cause negative impact the texture, flavor and colour of food product and cause decrease in protein digestibility, loss of amino acids and formation of toxic compounds (Vaishall et al. 2021, Yilma et al. 2023) Thus, acha is a source of many nutrients critical to human health (Jillian, 2020, Ijemi et al. 2025). Another noteworthy fonio nutritional benefit is its supply of nutrients, including iron (Fe), zinc(Zn) and phosphorus (P) which are important for facilitating cognitive function, bone health, high energy levels and many other functions (Jillian, 2020). Additionally, it contains some calcium, magnesium, potassium, manganese and folate. The high iron content of fonio, and also zinc, makes it particularly suitable for people following a vegan diet, since the diet can increase the risk for low iron intake and anemia since many iron-rich foods such as meat are avoided (Jillian, 2020). Within fonio you'll also find B vitamins, including thiamine, riboflavin, folate and niacin, which help support the conversion of nutrients into useable energy, support glucose metabolism, and play a key role in healthy muscle, nerve, heart and brain functions (Jillian, 2020).

One of the ingredients for bread-making is sugar (NAFDAC, 2023). Sugar is the primary food for the yeast, acts as a preservative like salt against mold and also help bread keep its moisture (Del Coro, 2022). Sugar contains high calories with no essential nutrients, thus, causing metabolic problems like type II diabetes and obesity (Ikechukwu et al., 2017). Substitution of sugar with date fruit pulp in bakery products has been advocated for as a means of curbing the afore-stated health related issues (Ikechukwu et al., 2017). Some natural alternatives of white sugar for bakery products are: raw honey, maple, syrup, molasses, corn syrup, steria, xylitol, agara, nectar, brown rice syrup, evaporated cane juice, black strap molasses, date sugar and organic sugar (Juntti, 2025).

Phoenix dactylifera is the type species of genus *Phoenix*, whose fruit develop through five different stages Hanabauk, Kimri, Khalal (or Bisr), Rutab, and Tamr and become edible in the final three stages as a result of decreased bitterness, increased sweetness, and improved tenderness, and succulence (Chafi et al., 2015). Date palm (*Phoenix dactylifera*) fruit is regarded as complete food due to its high nutritional content (Al-Mssallem, 2020). Atleast six vitamins (thiamine, riboflavin, niacin, ascorbic acid, pyridoxine and vitamin A) have been reported to be present in dates in visible consideration (Perveen et al., 2020; Rahman et al., 2022, Yasawy 2016). Dates are pitted, dried and then grind into a powder to make date sugar. Date sugar is a less refined sugar than typical white sugar and can be substituted in many foods and beverages (Sablani et al., 2008, Amerinasab et al., 2015). Juntti (2025) reported that when it comes to swapping date sugar for white sugar in recipes, date sugar can be substituted 1:1 ratio for white sugar. Date sugar is lower in glycemic index than cane or beet sugar (Conklin and Stilwell, 2007; Pasupuleti, 2008), honey, coconut sugar and any other alternatives (Shiza et al., 2022). it is the highest antioxidant sweetener (Babarinde et al., 2016). Like many other fruits, dates contain measurable levels of calcium oxalates (WebMD, 2023) and can be used as a practical supplement for iron deficiency without any side effects (Siavoshi et al., 2020, Wang et al., 2021, Safran et al., 2024). Date fruit is traditionally known as a tonic actor for pregnant women who are close to giving birth (Al-Kuran et al., 2011; Chiraz et al., 2020) and during their lactation period (Chiraz et al., 2020).

Bread is a staple food eaten by almost everyone. Wheat flour is one of the main raw material needed for its production alongside sugar but wheat is rich in carbohydrate, low in protein and contains gluten which can cause diabetes, celiac disease and has a negative effect for people with gluten intolerance thus diabetics are therefore afraid to eat normal wheat bread. A high consumption of refined sugar which is stripped of all nutrients (Kris 2012), is associated with high risk of diabetes, dental caries, asthma, liver, heart and kidney diseases, overweight, eczema and neurodevelopmental disorders in children and adults (Sharlene, 2021; Ikechukwu et al., 2017). This objective of the study was to ascertain the appropriate ratio that highly edible, nutritious and health - worth bread can be produced using wheat, acha and date fruit flour blend that can be consumed by everyone including diabetics while reducing incidences of celiac disease, gluten intolerance and protein energy malnutrition.

Materials and Methods

Procurement of materials

Wheat flour, Acha grain (*Digitaria exilis*), Date palm fruit (*Phoenix datylifera*) and the other ingredients like sugar, yeast, baking fat, baking powder, milk flavor and salt were all purchased from North Bank Market in Makurdi Benue State. All chemicals used were of analytical

Preparation of raw materials

Preparation of Acha flour

Acha flour was prepared according to the procedure reported by Olagunju et al. (2020) with slight modification. The Acha grains were washed with tap water to separate stones and sand, then, they were dried in the cabinet dryer at 50°C for 6 hours. The resultant dried Acha was milled and sieved into flour using the hammer mill with 0.5 mm screen size.

Preparation of Date fruit flour

The Date palm fruit flour (powder) was prepared according to Ikechukwu et al. (2017) with slight modification. The date palm fruit was de-seeded, sorted and washed. The sorted deseeded broken dates are then oven dried at 80°C for 2hours. The dried dates are milled then sieved to date palm powder. The date flour is then packaged in a sealed plastic and stored at 4°C.

Formulation of the composite flour with ingredients for bread making

The formulation of the composite flour with ingredients for bread making was presented in Table 1. The composite wheat-acha- date fruit flour was blended in the ratios of 80:10:10 as sample B, 70:20:10 as sample C and 60:30:10 as sample D. Sample A was 100:0:0 flour and it served as the control. The flours were thoroughly mixed to obtain a homogenous blend and stored at ambient temperature (30±2°C) in air tight container.

Production of bread

Bread was produced using the straight dough method involving bulk fermentation as reported by Olaoye and Obidegwe (2018). A quantity (100g) each of the flour samples were weighed and an addition of required amount of water and other ingredients was done to obtain dough and kneaded on a pastry-board to smoothen. The dough was initially fermented for 1 hour at 30°C before subsequently kneaded to expel carbon dioxide and then, it was tightened-up to ensure improvement in the textural properties of the bread. The dough was sized and molded into the baking pans for final proofing at 30°C for 2 hours. Baking of the dough was carried out in a forced air convection electric oven (380V, ROHS Deck Baking Oven, Hangzhou 311121, China) at 230°C for 30 minutes.

Ingredients	A	B	C	D
Wheat	100	80	70	60
Acha	-	10	20	30
Date	-	10	10	10
Sugar	10	-	-	-
Salt	1	1	1	1
Yeast	2	2	2	2
B/Fats	5	5	5	5
B/Powder	20	20	20	20
Milk Flavor	10	10	10	10

Source: Ayo et al. (2024) – Modified.

KEY: A - 100% Wheat flour (Control).

B - 80% Wheat Flour; 10% Acha Flour; 10% Date Flour.

C - 70% Wheat Flour; 20% Acha Flour; 10%Date Flour.

D - 60% Wheat Flour; 30% Acha Flour; 10% Date Flour.

Table 1: Formulation of the composite flour with ingredients for bread making.

Determination of amino acid profile

The Amino Acid profile in the samples were determined using methods described by Benitez (1989). The known sample was dried to constant weight, defatted, hydrolyzed, evaporated in a rotary evaporator and loaded into the Applied Biosystems PTH Amino Acid Analyzer.

Defatting Sample

The sample was defatted using chloroform/methanol mixture of ratio 2:1. About 5g of the sample was put in extraction thimble (or filter paper) and extracted for 15 hours in soxhlet extraction apparatus (AOAC, 2006).

Nitrogen determination

A small amount (150mg) of ground sample was weighed, wrapped in whatman filter paper (No.1) and put in the Kjeldhal digestion flask. Concentrated sulphuric acid (10ml) was added. Catalyst mixture (0.5g) containing sodium sulphate (Na_2SO_4), copper sulphate ($CuSO_4$) and selenium oxide (SeO_2) in the ratio of 10:5:1 was added into the flask to facilitate digestion. Six pieces of anti-bumping granules were added. The flask was then put in Kjeldhal digestion apparatus for 3 hours until the liquid turned light green. The digested sample was cooled and diluted with distilled water to 100ml in standard volumetric flask. Aliquot (10m1) of the diluted solution with 10ml of 45% sodium hydroxide was put into the Markham distillation apparatus and distilled into 10ml of 2% boric acid containing 4 drops of bromocresol green/methyl red indicator until about 70ml of distillate was collected. The distillate was then titrated with standardize 0.01 N hydrochloric acid to grey coloured end point.

$$\text{Percentage Nitrogen} = \frac{(a-b) \times 0.01 \times 14 \times V \times 100}{W \times C}$$

Where:

a: Titre value of the digested sample.
b: Titre value of blank sample.
V: Volume after dilution (100ml).
W: Weight of dried sample (mg).
C: Aliquot of the sample used (10ml).
14: Nitrogen constant in mg.

Hydrolysis of the sample

A known weight (mentioned in the calculation sheet) of the defatted sample was weighed into glass ampoule. 7ml of 6NHCL was added and oxygen was expelled by passing nitrogen into the ampoule (this is to avoid possible oxidation of some amino acids during hydrolysis example methionine and cystine). The glass ampoule was then sealed with Bunsen burner flame and put in an oven preset at 105°C± 5°C for 22 hours. The ampoule was allowed to cool before broken open at the tip and the content was filtered to remove the humins. It should be noted that tryptophan is destroyed by 6N HCl. During acidic hydrolysis, asparagine and glutamine are transformed into aspartic and glutamic acid respectively, and cysteine and methionine are partially oxidized (Peace and Gilani,2005). The filtrate was then evaporated to dryness using rotary evaporator. The residue was dissolved with 5ml to acetate buffer (pH 2.0) and stored in plastic specimen bottles, which were kept in the freezer.

Loading of the hydrolysate into analyzer

The amount loaded was 60microlitre. Norleucine (10 microlitre) was added to the sample and amino acid standard mixture. This was dispensed into the cartridge of the analyzer. The analyzer is designed to separate and analyze free acidic, neutral and basic amino acids of the hydrolysate.

Norleucine is an internal standard. It corrects any variation that may occur in the sample and amino acids standard mixture. An internal standard should be included with all samples, both standards mixture and sample being analysed. Norleucine is most commonly used for this purpose. Approximately 10 micromitre (0.025micro mole) of norleucine should be included with each hydrolysate sample and standard mixture. The exact amount of an internal standard is unimportant as long as exactly the same amount is used each time.

List of amino acids which will be separated sufficiently for accurate integration: Leucine, lysine, isoleucine, phenylalanine, tryptophan, valine, methionine, proline, arginine, tyrosine, histdine, cystine, alanine, glutamic acid, glycine, threonine, serine and aspartic acid.

Method of calculating amino acid values

An integrator attached to the Analyzer calculates the peak area proportional to the concentration of each of the amino acids.

Determination of vitamins composition of bread samples
Determination of vitamin composition

Vitamin A (β-Carotene) and vitamin C was determined by the method of (AOAC, 2005).

Determination of vitamin B1 (thiamine)

The thiamine content was determined using the scalar analyzer method of AOAC, (2015). Five grams (5 g) of each sample of composite four was homogenized in 5 ml normal ethanoic sodium hydroxide solution. The homogenate was filtered and made up to 100 ml with the extract solution. A 10ml aliquot of the extract was dispensed into a flask and 10ml of potassium dichromate solution added. The resultant solution was incubated for 15 minutes at room temperature (25°C±3°C). The absorption was read from the spectrophotometer at 360 nm using a reagent blank to standardize the instrument at zero. The thiamine content was calculated as follows:

$$Vitamin\ B1\ \left(\frac{mg}{100}\right) = +\frac{Sample\ absorbance\ x\ concentration\ x\ Dilution\ factor}{Concentration}$$

Determination of vitamin B2 (riboflavin)

The riboflavin content was determined according to AOAC (2015) method. Two grams (2 g) of composite four samples were placed in a conical flask and 50 ml of 0.2N HCl was added to the sample, boiled for 1 hour, and then cooled. The pH was adjusted to 6.0 using sodium hydroxide 1N HCl was added to the sample solution to lower the pH to 4.5. The solution was filtered into 100 ml measuring flask and made to volume with water. In order to remove interference, two tubes were taken, labeled 1 and 2. Ten milliliter of filtrate and 1ml of riboflavin standard were added to test tube 2. About 1 ml of glacial acetic acid was added to each tube and mixed, and then 0.5ml of 3% KMnO4 solution was added to each tube. They were allowed to stand for 2 minutes, after which 0.5ml of 3% H2SO4 was added and agitated. The fluorimeter was adjusted to excitation wavelength of 470 nm and emission wavelength of 525 nm. The fluorimeter was adjusted to zero deflection against 0.1N H2SO4 and 100 against tube 2 (standard). The fluorescence of tube 1 was read. Two milliliter of sodium hydrogen sulphate was added to both tubes and the fluorescence measured within 10 seconds. This was recorded as blank reading.

$$Vitamin\ B2\left(\frac{mg}{100g}\right) = \frac{Sample\ read - Blank\ read}{(sample\ read - blank\ read) - sample\ read + standard\ tube - sample\ read + standard\ blank} X \frac{1}{sample\ weight}$$

Determination of pyridoxine (vitamin B6) content

The determination of Vitamin B6 was performed using a BUCK Scientific HPLC system (Model: BLC-10/11) following AOAC (2012) methodology. The procedure involved combining 3.0 grams of each sample with 5 mL of n-hexane and 20 mL of HPLC grade water. This mixture underwent homogenization at 12000 rpm, followed by centrifugation at 3500 x g for 30 minutes. The resulting solution was filtered sequentially through Whatman No. 1 filter paper and a 0.45μm membrane. Subsequently, 15 microliters of the supernatant were injected into the HPLC system, which was equipped with a UV detector operating at 254nm. The vitamin content in the samples was quantified by comparing their peak areas with those of standard vitamin solutions.

Determination of vitamin B12 (Cobalamin) content

Vitamin B12 was also determined using AOAC, 2015.

Determination of mineral composition of the bread samples

Determination of Iron

The Iron content was determined following the method of Tivde et al., (2021). Five milliliters of digested sample of composite four was placed in a 50 ml volumetric flask. Then 3ml of phenanthroline solution, 2 ml of hydrochloric acid and 1ml of hydroxylamine solution were added to the sample in sequence. The sample solution was boiled for 2 minutes and 9 ml of ammonium acetate buffer solution was added to the solution. The solution was diluted with water to 50ml volume. The absorbance was determined at 510 nm wavelength. Iron standard solution was prepared in order to plot a calibration curve to determine the concentration of the sample. Standard solution containing 100 mg/ml of ferric irons was prepared from 1g pure iron wires. The wires were dissolved in 100 ml concentrated nitric acid, boiled in a water bath and diluted to 100 ml with distilled water after cooling. Standard solutions of known concentrations were prepared by pipetting 2, 4, 6, 8 and 10ml standard iron solution into 100ml volumetric flasks and made up to volume.

Determination of Phosphorus

The Phosphorus content was determined by a procedure described by Tivde et al., (2021). Using a flame photometer. Phosphorus standard was prepared. The standard solution was used to calibrate the instrument read out of composite four. The meter reading was at 100% E (emission) to aspire the top concentration of the standards. The %E of all the intermediate standard curves were plotted on linear graph paper with these readings. The sample solution was aspired on the instrument and the readings (% E) were recorded. The concentration of the element in the sample solution was read from the standard curve.

$$Phosphorus = \frac{(Ppm\ x\ 100\ x\ DF)}{1\ million}$$

Determination of Zinc

The Zinc content was determined by a procedure described by Tivde et al., (2021). Five milliliters (5 ml) of the test solution was pipetted into 50 ml graduated flask. Then 10 ml of molybdate mixture was added and diluted to mark with water. It was allowed to stand for 30 minutes for color development. The absorbance was measured at 660 nm against a blank. A curve relating absorbance to mg zinc present was constructed. Using the zinc standard solution, and following the same procedure for the test sample, a standard curve was plotted to determine the concentration of zinc in the composite flour sample.

$$Zinc = \frac{(Graph\ reading\ x\ solution\ volume)}{100}$$

Determination of Calcium and Potassium

Calcium and Potassium was determined by AOAC (2000) method using Atomic Absorption Spectrophotometer (AAS) (Perkin - Elmer - Crop, Norwalk model 560).

Statistical Analysis

Triplicate data obtained were subjected to statistical analysis using the Statistical Package for the Social Science (SPSS) software. The analysis of variance (ANOVA) were used to determine significance difference between the mean (P<0.05) while the means was separated using Duncan Multiple Range Test (DMRT).

Results and Discussion

Amino Acid Profile of the Bread Produced from Wheat, Acha and Date Palm Fruit Flour Blend

The Amino acid profile of the bread produced from the flour blends of wheat, acha and date fruit is presented in Table 2. Sample D (60% Wheat flour, 30% Acha flour, 10% Date fruit) had the highest value in leucine (8.71 mg/g protein), lysine (3.84 mg/g protein), isoleucine (3.52 mg/g protein), phenylalanine(4.55mg/g protein), tryptophan (1.28 mg/g protein), valine (5.41 mg/g protein), methionine (1.30 mg/g protein), proline (5.64 mg/g protein), arginine (4.85 mg/g protein), tyrosine (5.40 mg/g protein), histidine (2.30 mg/g protein), cysteine (2.03 mg/g protein), alanine (15.22 mg/g protein), and glutamic acid (15.22 mg/g protein) while sample A (100% Wheat flour - the control) had the lowest value in leucine (7.14 mg/g protein), lysine (3.14 mg/g protein), isoleucine (3.23 mg/g protein), phenylalanine (3.74 mg/g protein), tryptophan (1.20 mg/g protein), valine (4.63 mg/g protein), methionine (1.23 mg/g protein), proline (4.09 mg/g protein), arginine (3.83 mg/g protein), tyrosine (3.52 mg/g protein), histidine (2.15 mg/g protein) and cysteine (1.54 mg/g protein). Sample B (80% Wheat flour, 10% Acha flour, 10% Date fruit) had the highest value in glycine (3.51mg/g protein) and aspartic acid (6.02 mg/g protein) while sample D (60% Wheat flour, 30% Acha, 10% date fruit) had the lowest value (3.32 mg/g protein) and sample C (70% Wheat flour, 20% Acha flour, 10% Date fruit) had the lowest value (5.13 mg/g protein) for glycine and aspartic acid respectively. Sample A (100% Wheat flour - the control) has the highest value in alanine (4.36 mg/g protein) threonine (3.56 mg/g protein) and serine (4.22 mg/g protein) while sample C (70% Wheat, 20% Acha, 10% Date fruit) has the lowest value in alanine (3.78 mg/g protein), threonine (3.10 mg/g protein) and serine (3.68 mg/g protein). From the result, it was observed that there was an increase in amino acid as wheat flour was substituted with acha flour and date fruit flour (samples B- D). Sample D (60% Wheat flour, 30% Acha flour, 10% Date fruit) that had more quantity of acha flour had the highest value of about thirteen (13) amino acids with essential amino acids included. This result is in agreement with Ayo et al (2018), Chinwe (2015) and EFRT (2000) who reported at different occasion that acha is rich in amino acids. Ayo et al. (2018) reported that acha grains are rich in amino acids; leucine (9.8%), methionine (5.6%), valine (5.8%) and cysteine which are vital to human health but deficient in today's major cereals. Acha grain is reported to contain almost twice as much methionine as egg protein does (Temple and Bassa, 1991). Saleh et al. (2013), Thacker (1984) and Bhargava (2005) reported that the overall amino acid profile of acha was found to be as good as or even better compared to those of the traditionally used cereals like maize, millet, milo, wheat, barley and rye because the amino acid composition of hungry rice is not limited in the indispensable amino acids of methionine, tryptophan, phenylalanine (Ihekoronye and Ngoddy 1985). Al -Shahib and Marshall (2003) also reported that dates contains 23 types of amino acids (Siavoshi et al., 2020; Wang et al., 2021; Safran et al., 2024). This result is also in agreement with Ijemi et al. (2025) who reported acha to be rich in protein and amino acid is the building block of proteins (Michael and Shamin, 2024) thus confirming why sample D had more amino acids than the other samples.

Wheat is relatively low in quantity and quality in terms of protein content (Seal et al., 2021). Anjum (2005) and FAO (2013) reported that amino acid composition of wheat is quite unbalanced, lacking the essential amino acids like lysine and methionine. Processing wheat into various products further depletes it of essential amino acids (Anjum, 2005; FAO, 2013).

Amino Acids	A	B	C	D
Leucine	7.14	7.80	8.23	8.71
Lysine	3.14	3.26	3.31	3.83
Isoleucine	3.23	3.36	3.44	3.52
Phenylalanine	3.73	4.03	4.27	4.55
Tryptophan	1.20	1.21	1.22	1.28
Valine	4.63	4.93	5.34	5.41
Methionine	1.23	1.25	1.34	1.38
Proline	4.09	4.80	5.05	5.64
Arginine	3.83	4.00	4.49	4.85
Tyrosine	3.52	3.75	3.95	5.40
Histidine	2.15	2.21	2.23	2.30
Cystine	1.54	1.60	1.75	2.03
Alanine	4.36	4.11	3.78	4.03
Glutamic Acid	14.39	14.07	13.14	15.22
Glycine	3.45	3.51	3.45	3.32
Threonine	3.56	3.31	3.10	3.25
Serine	4.22	3.84	3.63	3.95
Aspartic Acid	5.88	6.02	5.13	5.57

KEY: A = Bread made from 100% Wheat flour (Control).

B = Bread made from 80% Wheat flour, 10% Acha flour and 10% Date flour.

C = Bread made from 70% Wheat flour, 20% Acha flour and 10% Date flour.

D = Bread made from 60% Wheat flour, 30% Acha flour and 10% of Date flour.

Table 2: Amino Acid Profile of the Bread Produced from Wheat, Acha and Date Palm Fruit Flour Blend.

Mineral Properties of the Bread

The mineral properties of the bread produced from Wheat flour, Acha flour and Date Fruit Flour blends are presented in Table 3. The minerals (Iron, Potassium, Phosphorus, Zinc and Calcium) increased with corresponding increase in the levels of substitution of Wheat flour with Acha flour and Date flour. The value of Iron ranged from 2.70 - 6.31mg/100g, Potassium ranged from 195.80 - 295.50mg/100g, Phosphorus ranged from 215.90 - 310.50 mg/100g, Zinc ranged from 1.50 - 3.39mg/100g and Calcium ranged from 90.59 - 110.30mg/100g with Sample A (100% Wheat flour) having the least values for all the minerals and Sample D (60% Wheat flour, 30% Acha flour, 10% Date flour) having the highest value for all the minerals. This result is in agreement with the result reported by Ayo et al. (2024) who reported an increase in phosphorus, iron and zinc as wheat flour was substituted with acha flour complemented with defatted Bambara groundnut flour. This result is also in agreemant with the report of Dabel et al. (2016) who reported an increase in calcium, iron and zinc as wheat flour was substituted with acha and mung beans flour. This result is also in agreement with Ijemi et al. (2025) who reported acha flour to have the highest value of ash content than wheat flour and later established that the more wheat flour was substituted with acha and date flour, the more the ash content increased. Ash content of any food is a measure of the total amount of minerals within the food product (Ojinnaka and Nnorom 2015, Hamza et al. 2014) and Chinma et al. (2012) reported that these minerals are capable of solving malnutrition problems.

Owheruo et al. (2023) reported that wheat is low in minerals while the National Academy Press (1996) reported Acha to be richer in Magnesium, Zinc and Manganese than any other cereal and The National Academies (2001) using a chart also reported Acha to be richer in Iron, Calcium, Phosphorus, Sodium, Potassium and Zinc than any other cereal. Jillian (2020) reported that another noteworthy fonio nutritional benefit is its supply of nutrients, including iron (Fe), zinc (Zn) and phosphorus (P) which are important for facilitating cognitive function, bone health, high energy levels and many other functions; additionally, it contains some calcium, magnesium, potassium, manganese and folate. The high iron content of fonio, and also zinc, makes it particularly suitable for people following a vegan diet, since the diet can increase the risk for low iron intake and anemia since many iron-rich foods such as meat are avoided (Jillian 2020). Dates also provides a wide range of essential nutrients and are very good source of dietary potassium and trace elements including boron, cobalt, copper, fluorine, magnesium, maganesse, selenium and zinc (Miller et al., 2002), a measurable levels of calcium oxalates (WebMD, 2023) and can be used as a practical supplement for iron deficiency without any side effects ((Siavoshi et al., 2020; Wang et al., 2021, Safran et al., 2024). These reports from Jillian, (2020), Miller et al (2002), Siavoshi et al. (2020), Wang et al. (2021), Safran et al. (2024) and WebMD (2023) would explain why Sample D (60% Wheat flour, 30% Acha flour and 10% Date Fruit) which contained more grams of Acha flour and contained Date fruit flour had the highest quantity of all the minerals (Iron, Potassium, Phosphorus, Zinc and Calcium) analyzed. It is also noteworthy to state that all the samples containing Acha flour and Date fruit flour (Sample B-D) also had more minerals than sample A, the control (100% Wheat flour).

Potassium is crucial to heart function and plays a key role in skeletal and smooth muscle contraction, making it important for normal digestive function (Chinwe et al., 2015). Kristeen Cherney (2024) reported that potassium assists in a range of essential body functions, including: blood pressure, normal water balance, muscle contractions, nerve impulses, digestion, heart rhythm and pH balance (acidity and alkalinity). Phosphorus is found mainly in bones and is a constituent of many vital compounds in the body, including ATP, DNA, phospholipids and the maintenance of buffers pH systems in the bodily fluids (Chinwe et al., 2015; Kobue - Lekalake et al., 2022). The main function of phosphorus is in the formation of bones and teeth; Calcium is necessary for teeth and bone health, whereas iron is crucial for the formation of heamoglobin, and plays an important role in the various metabolic processes (Kobue - Lekalake et al., 2022). Kobue - Lekalake et al. (2022) reported that the iron bioavailability from plant - based diets can range from 5 - 10%. Zinc aids in the growth and repair of tissues, boosts the immune system, and plays an important role in sperm survival (Gamonski, 2014, Chasapis et al., 2020).

Samples	Vitamin B1 (mg/100g)	Vitamin B2 (mg/100g)	Vitamin B6 (mg/100g)	Vitamin B12 (mg/100g)	Vitamin C (mg/100g)
A	$0.50^{d}\pm0.001$	$0.10^{d}\pm0.002$	$0.33^{c}\pm0.01$	$0.004^{c}\pm0.001$	$0.85^{c}\pm0.02$
B	$0.63^{c}\pm0.006$	$0.13^{c}\pm0.002$	$0.54^{b}\pm0.02$	$0.006^{bc}\pm0.00$	$0.93^{bc}\pm0.02$
C	$0.74^{b}\pm0.002$	$0.14^{b}\pm0.003$	$0.75^{a}\pm0.02$	$0.007^{b}\pm0.001$	$1.22^{b}\pm0.00$
D	$0.78^{a}\pm0.034$	$0.15^{a}\pm0.009$	$0.82^{a}\pm0.07$	$0.01^{a}\pm0.002$	$1.64^{a}\pm0.36$

Values are Means ± Standard Deviation of Triplicate Determination. Values followed by thesame subscript (s) within the same column are not significantly different at 5% probability level.

KEY: A - 100% Wheat flour (Control).

B - 80% Wheat Flour; 10% Acha Flour; 10% Date Flour.

C - 70% Wheat Flour; 20% Acha Flour; 10%Date Flour.

D - 60% Wheat Flour; 30% Acha Flour; 10% Date Flour.

Table 3: Mineral Composition of the Bread Produced from Wheat, Acha and Date Palm Fruit Flour Blend.

Vitamin Properties of the Bread

The Vitamin properties of the bread produced from Wheat flour, Acha flour and Date fruit flour blend are presented in Table 4. The vitamins (Vitamin B1, Vitamin B2, Vitamin B6, Vitamin B12 and Vitamin C) increased with corresponding increase in the levels of substitution of Wheat flour with Acha flour and Date flour from 0.5 - 0.78mg/100g, 0.10 - 0.15mg/100g, Vitamin B6 ranged from 0.33 - 0.82mg/100g, Vitamin B12 ranged from 0.004 - 0.01mg/100g and Vitamin C ranged from 0.85 - 1.64mg/100g with Sample A (100% Wheat flour) having the least values for all the vitamins and Sample D (60% Wheat flour, 30% Acha flour, 10% Date flour) having the highest value for all the vitamins. This result is in agreement with Dabel et al (2016) who reported an increase in vitamin as wheat flour was substituted with acha and mung beans. The result is also in agreement with Alebiosu (2024) who reported high values of vitamin for acha based complementary food.

Al-Hooti et al. (1995) reported that dates contain at least six vitamins: thiamine, riboflavin, niacin, ascorbic acid, pyridoxine and vitamin A (Perveen et al., 2020; Rahman et al., 2022; Yasawy 2016; Tamirat et al., 2024). Anjum (2005) and FAO (2013) reported that thiamine, riboflavin, niacin, and small amounts of vitamin A are present in wheat grain, but the milling processes removes most of these nutrients with the bran and germ. Owheruo et al. (2023), also reported that Wheat is low in vitamins. This explains why sample D (60% Wheat flour, 30% Acha flour and 10% date flour) had the highest value for all the vitamins (Vitamin B1, Vitamin B2, Vitamin B6, Vitamin B12 and Vitamin C) analyzed while sample A (100% Wheat flour) had the lowest value. There was no significant difference in sample C (70% Wheat flour, 20% Acha flour and 10% date flour) and sample D (60% Wheat flour, 30% Acha flour and 10% date flour) for vitamin B6. It is also noteworthy to state that all the samples that had Acha flour and Date fruit flour (Sample B - D) had higher values than sample A that had no Acha flour nor Date fruit flour. Vitamin B1 functions as the coenzyme thiamine pyrophosphate in the metabolism of carbohydrates and branched- chain amino acid and is essential for glucose metabolism and nerve, muscle, and heart function (Brazier, 2023). Riboflavin helps support growth and development, energy production, including the breakdown of fats, proteins, and carbohydrates into energy (Hobbs, 2023). Vitamin B6 has been widely studied for its role in disease prevention. Adequate blood levels of B6 may be associated with lower risk of cancers, compared to low blood levels (Harvard University, 2024). Vitamin C is vital for connective tissue, bone, and tooth health (Hill and Kelly, 2024). The B vitamins are needed for carbohydrate and protein metabolism, and are essential for growth, well structuring and functioning of the cells (Ahure et al., 2020).

Samples	Iron (mg/100g)	Potassium (mg/100g)	Phosphorus (mg/100g)	Zinc (mg/100g)	Calcium (mg/100g)
A	$2.70^{d}\pm0.09$	$195.80^{d}\pm0.05$	$215.90^{d}\pm0.00$	$1.50^{d}\pm0.10$	$90.59^{d}\pm0.08$
B	$3.57^{c}\pm0.02$	$212.50^{c}\pm1.76$	$231.00^{c}\pm0.00$	$1.90^{c}\pm0.04$	$92.41^{c}\pm0.09$
C	$4.21^{b}\pm0.01$	$242.60^{b}\pm0.02$	$254.20^{b}\pm0.05$	$2.30^{b}\pm0.04$	$96.28^{b}\pm0.06$
D	$6.31^{a}\pm0.03$	$295.50^{a}\pm0.09$	$310.50^{a}\pm0.16$	$3.39^{a}\pm0.03$	$110.3^{a}\pm0.02$

Values are Means ± Standard Deviation of Triplicate Determination. Values followed by thesame subscript (s) within the same column are not significantly different at 5% probability level.

KEY: A - 100% Wheat flour (Control).

B - 80% Wheat Flour; 10% Acha Flour; 10% Date Flour.

C - 70% Wheat Flour; 20% Acha Flour; 10%Date Flour.

D - 60% Wheat Flour; 30% Acha Flour; 10% Date Flour.

Table 4: Vitamin Composition of the Bread Produced from Wheat, Acha and Date Palm Fruit Flour Blend.

Conclusion

This study assessed the amino acid profile and nutritional properties of bread made from the flour blends of wheat, acha and date palm fruit. The study discovered that sample D had about 13 amino acids including amino acids that prevents denaturation, it was also more nutritious in terms of vitamins and minerals that can combat cases of malnutrition and are necessary to fight off diseases. Sample

D is therefore recommended for the baking industry.

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