



Amino Acid Profile of Heat-processed *Canarium schweinfurthii* Pulp

Ernest A. Anyalogbu^{1*}, Eugene N. Onyeike² and Michael O. Monanu²

¹Department of Biotechnology, Federal University of Technology, Owerri, Nigeria.

²Department of Biochemistry, Nutrition and Toxicology Unit, Faculty of Science, College of Natural and Applied Sciences, University of Port Harcourt, Port Harcourt, Nigeria.

Authors' contributions

This work was carried out in collaboration between all authors. All authors read and approved the final manuscript.

Original Research Article

Received 21st March 2014
Accepted 12th May 2014
Published 19th June 2014

ABSTRACT

Amino acid composition of a plant food is an indicator of its protein quality. This could be altered by the processing method. Processed fruit of *Canarium schweinfurthii*, a *Bursereceae*, is common traditional snacking item in Nigeria. The pulp of raw and macerated samples of *C. schweinfurthii* were dried, ground into powders and analyzed for protein and amino acid contents using standard methods. The pulp of raw and macerated samples contain all the amino acids found naturally in plant protein. Glu (6.72-9.03g/100g protein), a non-essential amino acid was the most abundant amino acid followed by Leu (5.35-6.21g/100g protein) an essential amino acid. The concentrations of Glu, Gly, Ala, Cys, and Tyr and those of all the essential amino acids were increased by macerating the sample for 15 to 45min while others decreased. Peak values (on g/100g protein basis) were obtained for Lys(3.10), Thr(3.00), Val(3.66), Met(0.89), Ile(3.08), Phe(3.30), Glu(9.03), Gly(3.14), Ala(2.86), Cys(0.79), and Tyr(2.74) at 30min maceration (CS₃₀). The sample processed to accepted eaten tenderness (CS₃₀) on g/100g protein basis also recorded the highest values for the protein quality parameters: total amino acid(56.05), total essential amino acid with His(28.5), total non-essential amino acid(27.8), total neutral amino acid(33.4), total acidic amino acid(14.4), total sulphur amino acid(1.68) and total aromatic amino acid(6.04). Based on whole hen's egg and 1957 FAO provisional amino acid patterns, Met (0.57-0.89g/100g protein) scored lowest to become the first limiting amino acid in the sample. The plant food has potential as a source of high quality dietary protein.

*Corresponding author: E-mail: anyalogbu@yahoo.com;

Keywords: Maceration; snaking; dietary protein; limiting amino acids; pulp.

1. INTRODUCTION

The major challenge to food security in Nigeria and indeed the entire developing countries of the world is how to augment the shortage of protein in diets of a large section of its population [1,2,3]. Ekop [4] pointed out that part of the problem was insufficient protein of good quality. According to Kader et al. [5] and, Agbaire and Emoyan [6], fruits and seeds are rich in protein, oil, energy, minerals and vitamins. *Canarium schweinfurthii* (African elemi or bush candle), a *Burseraceae* is an ever green tree indigenous to West and Central Africa. It is locally known in Nigeria as; 'Ube mgba' (Igbo), 'Atili' (Hausa) or 'Origbo' (Yoruba). As shown in Plate 1, the fruit is a drupe containing a single triangular shaped seed surrounded by a delicious purplish green pulp [7]. *C. schweinfurthii* seed is macerated in hot water and the pulp eaten as snacking item in most parts of Nigeria.



Plate 1. *Canarium schweinfurthii* fruits

The value of food and feeding-stuff for protein nutrition depends primarily on its amino acid contents [2,4,8]. Nine of these amino acids are considered essential for humans as their body cannot synthesize them from other compounds and must be taken ready-made as part of the diet [9]. Earlier, Seligson and Mackey [10] reported that a dietary requirement for protein is a requirement for non-essential amino nitrogen and essential amino acids. Pisarikova [3] stated that high content of essential amino acids in plant food predetermines its use as a substitution for meat and bone meals.

This study is intended to determine the amino acid contents of raw and macerated *Canarium schweinfurthii* seed pulps as index of their protein quality.

2. MATERIALS AND METHODS

Wholesome fresh fruits of *Canarium schweinfurthii* purchased from Ngwa road market in Aba, Aba South Local Government Area of Abia State, Nigeria were washed in several changes of distilled water and divided into four (4) lots. The first lot was used raw and therefore labeled CS_{raw}. Trial processing showed that eaten tenderness was obtained by macerating the CS fruits in hot water (55°C) for 30min. The 2nd, 3rd and 4th lots were macerated (by putting in hot water flask, adding sufficient water (55°C) to cover them and the flask covered) for 15, 30 and 45 min and labelled CS₁₅, CS₃₀, and CS₄₅ respectively. The water was drained off and the fruit pulps scrapped from the stones, sliced thinly with a knife and dried for 48hr in an air-circulatory oven (50°C) (Model OVE.100.130M. Gallenkamp, UK). The oven-dried samples were ground in a mill (Model BL357. Kenwood, Birmingham UK), passed through a 60-mesh size screen and used in the analyses.

2.1 Crude Protein Determination

Crude protein was estimated (in triplicates) by a process developed by Johan Kjeldahl in 1883 as described in the Official Methods of Analysis (No. 2.057) of AOAC [11] in which Kjeldahl nitrogen was determined. The method involved the digestion of 2.0g sample with 20ml concentrated H₂SO₄, distillation of the digest to liberate ammonia which was trapped into 2.0% boric acid solution, followed by titration with 0.1NHCl. The % nitrogen was multiplied by a factor of 6.25 to obtain % protein, since protein, on the average contain 16% nitrogen [2].

2.2 Determination of Amino Acid Profile

The free acidic, neutral and basic amino acids resulting from hydrolysis of defatted stock sample were separated by ion-exchange chromatograph (IEC) with post column ninhydrin-based detection [12,13] using the Technicon Sequential Multisample (TSM) Amino Acid Analyzer (Technicon Instruments Corporation, New York).

2.3 Procedure

2.3.1 Defatting of sample

Five grams of the sample was weighed into the extraction thimble. The fat was extracted for 6hr with chloroform/methanol (2:1 v/v) mixture using Soxhlet extraction apparatus as described by AOAC [11].

2.3.2 Acid hydrolysis of defatted sample

The defatted sample (1.4705g) was weighed into a glass ampoule and 7ml of 6NHCl added. Oxygen was expelled from the ampoule by flushing with Nitrogen. This was to prevent possible oxidation of some amino acids such as methionine and cysteine during hydrolysis [14]. The glass ampoule was sealed with Bunsen burner flame and incubated in an oven (105°C±5°C) (Model OVE.100.130M. Gallenkamp, UK) for 22 hr to effect hydrolysis [15].

The ampoule was allowed 1 hr to cool to room temperature (29±1°C) before it was opened at the tip and the content filtered through Whatman Number 52 filter paper to remove humans [2,6].

The hydrolysate was evaporated to dryness at 40°C under vacuum in a rotary evaporator (Buchi Rotavapour, Switzerland) and the residue (free acidic, neutral and basic amino acids) dissolved with 5ml acetate buffer (pH 2.0). The acetate buffer solution was put in plastic specimen bottle and stored in the freezer (−4°C).

2.3.3 Ion-exchange chromatography

With a clean microsyringe (World Precision Instruments^R, UK), 10ml of the hydrolysate was collected and dispensed into the cartridge of the Technicon Sequential Multisample (TSM) Amino acid Analyzer. The analyzer is sodium-based cation-exchange chromatograph with post column ninhydrin derivatization. The amino acids were separated on the ion-exchange column through a combination of changes in pH and cation strength [17]. A temperature gradient in the column enhanced the separation.

The post column reaction between ninhydrin and amino acids eluted from the column formed Ruhemann's purple, a diketohydrindylidene-diketohydrindamine [18]. The reaction was monitored at 440nm and 570nm wavelengths. The period of analysis was 76min for each sample. The gas flow rate was 0.58ml/min at 60°C with reproducibility consistent within ±3%. Norleucine was added as internal standard and a standard mixture of amino acids was also analyzed under the same condition as the sample. The net height of each peak in the chromatogram produced by the chart recorder of the TSM (each representing an amino acid in the sample) was measured and the peak area calculated. The amino acids present in the sample were identified by matching their peak retention time in the chromatogram with those of the amino acids in the standard chromatogram. The concentration of each amino acid was calculated in g/100g protein from the peak area.

2.4 Determination of Protein Quality Parameters

The amino acid score for the essential amino acids was calculated using the FAO/WHO [19] formula as adapted by Onyeike et al [2]:

$$\text{Amino acid score} = \frac{\text{Amount of amino acid per sample protein (mg/g)}}{\text{Amount of amino acid per protein in reference pattern (mg/g)}} \times 100$$

The total amino acid (TAA), total essential amino acid (TEAA), TEAA as percentage of TAA (%TEAA), total neutral amino acid (TNAA), total acidic amino acid (TAAA), total basic amino acid (TBAA), total sulphur amino acid (TSAA), percentage cysteine in TSAA (%Cys/TSAA), total aromatic amino acid (TArAA), Leu/Ile ratio etc. were estimated from the amino acid profile. The predicted protein efficiency ratio (P-PER) was determined using one of the equations derived by Alsmeyer et al [20], as adapted by Adeyeye [21]:

$$\text{P-PER} = -0.468 + 0.454 (\text{Leu}) - 0.105 (\text{Tyr}).$$

2.5 Statistical Analysis

Data on crude protein were analyzed by analysis of variance (ANOVA) and means were compared by the Duncan's multiple range tests [22]. Significance was accepted at 5% level ($p \leq 0.05$) or 95% confidence limit.

3. RESULTS AND DISCUSSION

The essential and non-essential amino acid compositions of raw and macerated *C. schweinfurthii* (CS) pulp are shown in Tables 1 and 2 respectively. The pulp of raw and macerated samples was found to contain all the amino acids found naturally in plant protein. The concentration of all the amino acids was affected by the processing method.

Table 1. Essential amino acid concentrations^a (g/100g protein) of raw and macerated *C. schweinfurthii* pulp

Amino acid (in three letter language)	Sample			
	CS _{raw}	CS ₁₅	CS ₃₀	CS ₄₅
Lys	2.94	3.01	3.10	2.60
His	1.50	1.63	1.69	2.07
Arg	3.05	3.15	3.49	4.08
Thr	1.88	2.25	3.00	2.44
Val	2.90	3.49	3.66	3.37
Met	0.57	0.63	0.89	0.73
Ile	2.51	2.69	3.08	2.79
Leu	5.85	6.21	6.04	5.35
Phe	2.62	2.86	3.30	3.04

^aValues are means of duplicate determinations

Table 2. Non-essential amino acid concentrations^a (g/100g protein) of raw and macerated *C. schweinfurthii* pulp

Amino acid (in three letter languages)	Sample			
	CS _{raw}	CS ₁₅	CS ₃₀	CS ₄₅
Asp	6.89	6.36	5.34	4.36
Ser	2.16	2.07	1.89	1.48
Glu	7.14	7.22	9.03	6.72
Pro	2.23	2.12	2.01	1.91
Gly	1.99	2.24	3.14	2.14
Ala	1.70	2.77	2.86	1.47
Cys	0.66	0.90	0.79	0.66
Tyr	2.09	2.41	2.74	2.58

^aValues are means of duplicate determinations

As shown in Table 1, Leu (5.35-6.21g/100g protein) was the most abundant essential amino acid in the sample followed by Arg (3.05-4.08g/100g protein). This observation corroborate the reports of Olaofe et al. [23], Adeyeye [21], Aremu et al. [24], Aremu et al. [25] and Adeola [26] that Leu is the most abundant essential amino acid in Nigerian plant foods. The abundance of Leu in the test sample is of nutritional significance as together with Val and Ile, it constitutes the branch chain amino acids (BCAAs). BCAAs are found in high concentrations primarily in the skeletal muscle tissue. While Ile serves mainly as antioxidant, Leu and Val stimulate protein synthesis, suppress protein catabolism and serve as important fuel sources for skeletal muscle during period of extreme exertion such as exercise [27,28,29,30,26]. With the values of 2.9-3.66g/100g protein and 2.51-3.08g/100g protein obtained for Val and Ile respectively in this work, *C. schweinfurthii* pulp could be relatively adjudged rich in BCAAs. The second largest essential amino acid in the sample was Arg

(3.05-4.08g/100g protein). Arg is the precursor for the production of nitric oxide (NO). According to Alvares et al. [31], NO promote vasodilation in active muscle during exercise thereby improving strength, power and recovery. Consequently, the consumption of the sample by the sporting community is highly recommended for improved performance.

The most predominant amino acids in the sample were Glu (6.72-9.03g/100g protein) and Asp (6.86-4.36g/100g protein) - non-essential acidic amino acids. Earlier, Food and Agriculture Organization of the United Nations [32] reported Glu (18.80g/100g protein) as the most predominant amino acid in the sample (seed). The predominance of Glu and Asp have also been reported in some other plant foods: Soybeans [32], *Trigonella foenumgraceum* [33], yam (*Dioscorea dumetorum*) [34], groundnut seeds [21], peas (*P. sativum*) [8] and tamarind (*Tamarindus indica* L) [26]. The abundance of these acidic amino acids in the plant food is of significance. Glu is the source of alpha-amino group in the biosynthesis of most dispensable amino acids. Asp on the other hand is a precursor of the essential amino acids like Asn, Met, Thr and Lys [35]. Lys (2.64–3.10g/100g protein) and Phe (2.62-3.30g/100g protein) are other essential amino acids of note in the plant food. The observed value for Lys (2.94mg/100g protein) is slightly higher than the value (2.90mg/100g protein) recorded by FAO [32] for the sample. Lys according to Onyeike, et al [2] is usually limiting in most cereals and legumes. These food items are staple foods in most part of Nigeria where *C. schwenfurthii* pulp is commonly consumed as traditional snack. The high content of Lys in the plant food is therefore of nutritional significance since it would make good its deficiency in these staple foods. Apart from Lys, the values obtained for the amino acids (essential and non-essential) are lower than the corresponding values obtained by FAO [32] for the sample.

Tables 3 and 4 show the percentage changes in the concentrations of essential and non-essential amino acids of the sample when subjected to different maceration time. The result indicated that maceration had mainly increasing effect on the concentration of the all the essential amino acids and, Glu, Gly, Ala, Cys and Tyr (non-essential amino acids). Peak values (on g/100g protein basis) were obtained for Leu(6.21) at 15min maceration; Lys(3.1), Thr(3.00), Val(3.66), Met(0.89), Ile(3.08), Phe(3.30) at 30min maceration and; His(2.07) and Arg(4.08) at 45min maceration. The non-essential amino acids, Asp, Ser and Pro were progressively decreased as maceration time was increased from 15min to 45min giving the range; 4.36-6.89, 1.48-2.16 and 1.91-2.23g/100g protein (i.e. 7.29-36.4, 4.17-31.50 and 4.93-14.4% reduction) respectively.

Table 5 shows the crude protein contents and protein quality parameters of raw and macerated *C. schweinfurthii* samples. The raw sample recorded crude protein content of 14.6 ± 0.51 g/100g sample. This was lower than the value 20.43g/100g sample earlier obtained by Onimawo and Adukwu [36] for the same sample. The observed value (14.6 ± 0.51 g/100g sample) was significantly ($p \leq 0.05$) affected being progressively reduced to 12.56 ± 1.10 g/100g sample as maceration time was extended from 15min to 45min. Food processing methods have been reported to alter the nutrient contents of plant-based diets [37]. Earlier, Sefa-Dedeh et al. [38] stated that heat-processing (e.g. maceration) was a hydrothermal process that involved hydration and heating. In 2007, Hotz and Gibson [39] reported that heat disrupts the cell structure and membrane partitions of food plants, releasing nutrients from entrapment in the plant matrix. The observed increase in the amino acid of the sample with increased maceration time could be attributable to the “releasing effect” of heat on nutrients. It could also be as a result of inactivation of anti-nutritional factors that made susceptible nutrients unavailable. According to Yadav and Sehgal [40] and, Akinyele and Oroluntoba [41], thermal processing improves nutrient availability of foods by destroying certain antinutritional factors. The reduction noticed in both protein and some

amino acid contents could be attributable to the leaching of the nutrient into the processing water and possibly to a lesser extent (because of the low temperature (55°C) to Maillard reaction [42,21]. The leaching may have been facilitated by heat which was reported to increase the solubility of nutrients in the processing water [43].

Table 3. Difference in the essential amino acid concentrations (g/100g crude protein) between raw and macerated *C. schweinfurthii* pulp

Amino acid (in three letter language)	Difference (percentage difference) ^a		
	CS ₁₅ – CS _{raw}	CS ₃₀ – CS _{raw}	CS ₄₅ – CS _{raw}
Lys	0.07(2.38)	0.16(5.44)	-0.34(11.70)
His	0.13(8.67)	0.19(12.70)	0.57(38.0)
Arg	0.10(3.28)	0.44(14.40)	1.03(33.40)
Thr	0.37(19.70)	1.12(59.60)	0.56(29.80)
Val	0.59(20.40)	0.76(26.20)	0.47(16.20)
Met	0.06(10.50)	0.32(56.10)	0.16(28.10)
Ile	0.18(7.17)	0.57(22.70)	0.28(11.20)
Leu	0.36(6.15)	0.19(3.25)	-0.50(8.55)
Phe	0.24(9.16)	0.68(26.0)	0.42(16.0)

^aBracketed values are percentage changes in the amino acid level after heat-processing

Table 4. Differences in the non-essential amino acid concentrations (g/100g crude protein) between raw and macerated *C. schweinfurthii* pulp

Amino acids	Difference (percentage difference) ^a		
	CS ₁₅ – CS _{raw}	CS ₃₀ – CS _{raw}	CS ₄₅ – CS _{raw}
Asp	-0.53(7.69)	-1.55(22.50)	-2.53(36.72)
Ser	-0.09(4.17)	-0.27(12.50)	-0.68(31.50)
Glu	0.08(1.12)	1.89(26.50)	-0.42(5.88)
Pro	-0.11(4.93)	-0.22(9.87)	-0.32(14.40)
Gly	0.25(12.60)	1.15(57.80)	0.15(7.54)
Ala	1.07(62.90)	1.16(68.20)	-0.23(15.50)
Cys	0.24(36.36)	0.13(19.70)	0.0
Tyr	-0.32(15.30)	0.65(31.10)	0.49(23.5)

^aBracketed values are percentage changes in the amino acid level after heat-processing

The recommended daily allowance (RDA) of protein for men and women are 56 and 46g respectively [44], and 23-36g for children [45]. A 100g of the sample could therefore provide 22.5-26.1% and 27.3-31.7% of the RDA of protein for men and women respectively, and 42.6- 49.5% for children. The observed levels of crude protein (12.56±1.10-14.6±0.51g/100g sample) may qualify the sample as good source of dietary protein if bio-available and easily digestible by the body. The pulp of samples processed to acceptable eaten tenderness, i.e. CS₃₀, showed on the basis of g/100g protein the highest total amino acid (56.05), total essential amino acid with His (28.25), total non-essential amino acid (27.8), total neutral amino acid (33.4), total acidic amino acid (14.4), total sulphur amino acid (1.68) as well as total aromatic amino acid (6.04). Analyses of the data showed that the percentage ratio of TEAA to TAA (%TEAA) was high and increased progressively with processing time given the range 49.0-55.4%. This indicates that *C. schweinfurthii* pulp is a good source for EAAs which were made more available with processing. The TEAA value at every level of processing was higher than the 39%, 26% and 11% considered to be adequate for ideal protein food for infants, children and adults respectively [46,47]. The observed %TEAA values (49.0-55.4%)

could be favourably compared with those of proteins of animal sources; 54.8% in trunk fish [48], 50% in whole hen's egg [49], 43.7% in wing termites [50]. On pair wise comparisons, the effect of processing on the total neutral amino acid (27.2-33.4g/100g protein), total acidic amino acid (11.1-14.4g/100g protein) and total basic amino acid (7.49-8.75g/100g protein) were not pronounced. The percentage neutral amino acid (%TNA) ranged from 55.8 to 59.6 and was more than the %TAAA (23.2-28.8) and %TBAA (15.0-18.3) added together. This indicates that neutral amino acids formed the bulk of the amino acids in the sample. The total aromatic amino acids (TArAA) in the sample varied with processing time given the range 4.71-6.04g/100g protein. ArAAs (Phe and Tyr) are precursors of many important biological compound such neurotransmitters and hormones in the human body [51].

Table 5. Crude protein, P-PER, concentrations of essential, non-essential, neutral, acidic, basic, sulphur, aromatic amino acids etc (g/100g crude protein) of raw and heat-processed *C. schweinfurthii* pulp

Quality parameter	CS _{raw}	CS ₁₅	CS ₃₀	CS ₄₅
Protein (g/100g sample)	14.60±0.51 ^a	13.20±1.00 ^b	12.59±0.87 ^b	12.56±1.10 ^b
Total amino acid (TAA)	48.70	51.80	56.10	47.80
Total non-essential amino acid (TNEAA)	24.80	25.90	27.80	21.30
%TNEAA	51.0	50.0	49.60	44.60
Total essential amino acid(TEAA)				
*with His	23.80	25.90	28.30	26.50
*Without His	22.30	24.30	26.60	24.40
% TEAA with His	49.0	50.0	50.40	55.40
% TEAA without His	45.90	46.90	47.40	51.10
Total neutral amino acid (TNA)	27.20	30.50	33.40	28.0
% TNA	55.80	58.80	59.60	58.50
Total acidic amino acid (TAAA)	14.0	13.60	14.40	11.10
% TAAA	28.80	26.20	25.60	23.20
Total basic amino Acid (TBAA)	7.49	7.79	8.28	8.75
% TBAA	15.40	15.0	14.80	18.30
Total sulphur amino acid (TSAA)	1.23	1.36	1.68	1.39
% TSAA	2.53	2.62	3.0	2.91
% Cys in TSAA	53.70	53.70	47.0	47.50
Total aromatic amino acid (TArAA)	4.71	5.27	6.04	5.62
% TArAA	9.68	10.20	10.80	11.80
P-PER ^a	1.97	2.10	1.98	1.69
Leu/Ile ratio	2.33	2.31	1.96	1.92
Leu – Ile (difference)	3.34	3.52	2.96	2.56
% Leu-Ile	57.10	56.7	49.0	47.90

^aPredicted Protein Efficiency Ratio. Crude protein values bearing the same superscript letters are not significantly different at the 5% level ($p \geq 0.05$)

They are involved in the synthesis of the pigment melanin in hair, eye and skin [48]. The observed TArAA range (4.71-6.04g/100g protein) in the sample is lower than the range 6.8-11.8g/100g protein suggested for infant protein [46].

The predicted protein efficiency ratio (P-PER) of the sample ranged from 1.69 to 2.10. This is higher than 1.50, below which a protein is taken to be of low or poor quality [52]. The values though lower than 2.62 and 2.5 obtained for groundnut seed [21] and Casein [46] respectively, compared favourably with 1.21 (Cowpea) and 1.82 (Pigeon pea) [53], and 1.62 (Millet ogi) [54]. The Leu/Ile ratio of the sample decreased progressively with processing time given the range 2.33-1.92 i.e. 2.33:1-1.92:1. The values were comparable with the ideal

ratio of 1.8:1 suggested by FAO/WHO/UNU [46]. This moderate ratio would permit full use of the Lys and Ile content of the sample as excess Leu in foods was reported to interfere with their utilization [55].

The results show that Met (0.57-0.89/100g protein) and Cys (0.66-0.79g/100g protein) were the 1st and 2nd limiting amino acids respectively (Table 1 and 2). These two amino acids constitute the total sulphur amino acids (TSAA). The TSAA of the sample varied with processing time given the range 1.23-1.68g/100g protein. This is lower than the value 5.8g/100g protein recommended for infant [46]. According to Adeyeye and Aremu [56] most animal proteins are low in Cys while many vegetable proteins contain substantially more Cys than Met. The %Cys in TSAA obtained (47.02-53.7) compared favourably with 50.5% recorded for *Anacardium occidentale* [57]. The percentage of Cys in TSAA had been set at 50% in rat, chick and pig diets [13]. The sulphur amino acids could function as anti- or pro-oxidants depending on the type of oxidant stress and physiological circumstances [58]. Cys bonds with other protein in the hair shaft and increases the diameter of the hair shaft. Met on the other hand is needed for the synthesis of choline which forms lecithin and other phospholipids in the body [56]. It breaks down fats that could block arteries, increasing blood flow and consequently nutrient supply, to the hair follicle [59]. Cys also has positive effects on mineral absorption especially zinc [60].

Tables 6-7 show the essential amino acid scores of the raw and macerated *C. schweinfurthii* pulps based on whole hen's egg and FAO [61] provisional amino acid patterns. Essential amino acid score (percentage adequacy) as stated by Onyeike et al [2] is a grading in which the quality of a protein can be established by comparing its amino acid contents with that of a reference protein and it recognizes that the value of a protein is determined by the levels of its essential amino acid. Relative to the hen's egg amino acid pattern, His scored highest (61.7-85.1%) followed by Leu (60.7-70.4%) and then Arg (50.0-60.9%). The food plant could supply as much as 50% of whole hen's egg content of Thr only at CS₃₀; Val at CS₁₅ and CS₃₀; and Phe at CS₃₀ and CS₄₅. Except for Met that scored 24.7-38.6%, *C. schweinfurthii* pulp at all levels of processing showed good comparisons with FAO provisional amino acid pattern. In both reference patterns, Met scored lowest re-emphasizing its position as the first limiting amino acid at all levels of processing.

Table 6. Essential amino acid scores of raw and heat-processed *C. schweinfurthii* pulp based on whole hen's egg amino acids^a

Essential amino acid	Hen's egg (mg amino acid/g Total N)	Chemical score (%)			
		CS _{raw}	CS ₁₅	CS ₃₀	CS ₄₅
Lys	436	42.2	43.1	44.5	37.3
His	152	61.7	67.0	69.5	85.1
Thr	320	36.7	43.9	58.6	47.7
Arg	381	50.0	51.7	57.2	66.9
Val	428	42.4	51.0	53.5	49.2
Met	210	17.0	18.8	26.5	21.7
Met+Cys	362	21.2	23.5	29.0	24.0
Ile	393	39.9	42.8	49.0	44.3
Leu	551	66.4	70.4	68.5	60.7
Phe	358	45.8	49.9	57.6	53.1
Phe +Tyr	618	47.6	53.3	61.1	56.9

^aFAO,1970

This means that if the food plant is the sole protein source for an individual, to correct for the amino acid needs from any of the samples it becomes necessary that 100/17 or 5.9times as much CS_{raw} protein; 100/18.8 or 5.3times as much as CS₁₅ protein; 100/26.5 or 3.8times as much as CS₃₀ protein and; 100/21.7 or 4.6times as much CS₄₅ protein would have to be taken [62].

Table 7. Essential amino acid scores of raw and heat-processed *C. schweinfurthii* pulp based on FAO amino acid provisional pattern^a

Essential amino acid	FAO provisional pattern (mg amino acid/g total N)	Chemical score (%)			
		CS _{raw}	CS ₁₅	CS ₃₀	CS ₄₅
Lys	270	68.1	69.7	71.8	60.2
His	87	107.8	117.2	121.4	148.7
Thr	180	65.3	78.2	104.2	84.7
Val	270	67.2	80.8	84.7	78.0
Met	144	24.7	27.4	38.6	31.7
Met+Cys	270	28.5	31.5	38.9	32.2
Ile	270	58.1	62.3	71.3	64.6
Leu	306	109.5	126.8	123.4	109.3
Phe	180	91.0	99.3	114.6	105.6
Phe +Tyr	360	81.8	91.5	104.9	97.6

^aFAO 1957

4. CONCLUSION

C. schweinfurthii pulp at all levels of processing especially at CS₃₀ is comparatively rich in BCAAs and Arg which make the consumption invaluable in improving performance in sports. Relative to established standards, most of the protein quality parameters of the plant food were outstanding—indicative of the high potential of the plant food as a source of good quality dietary protein.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

REFERENCES

1. Akaninwor JO, Abbey BW, Ayalogu EO. Profile of protein-energy malnutrition amongst children under four years in urban areas of Rivers State. West African Journal of Medicine. 1996;15:50-55.
2. Onyeike EN, Ayalogu EO, Okaraonye CC. Nutritive value of the larvae of raphia palm beetles (*Oryctes rhinoceros*) and weevil (*Rhyncophorus pheonicis*). Journal of the Science of Food and Agriculture. 2005;85:1822-1828.
3. Pisarikova B, Kracmar S, Herzig I. Amino acid contents and biological value of protein in various amaranth species. Czech Journal of Animal Science. 2005;50(4):169-174.
4. Ekop AS. Changes in amino acid composition of African yam beans (*Sphenostylis stenocarpas*) and African locust beans (*Parkia filicoida*) on cooking. Pakistan Journal of Nutrition. 2006;5(3):254-256.
5. Kader A, Perkins-Veazie PM, Lester GE. Nutritional quality of fruits, nuts and vegetables and their importance in human health. USDA Hand Book. 2004;66.

6. Agbaire PO, Emoyan OO. Nutritional and antinutritional levels of some local vegetables from Delta State, Nigeria. *African Journal Food Science*. 2012;6(1):8-11.
7. Agu HO, Ukonze JA, Uchola NO. Quality characteristics of crude and refined atili oil. *Pakistan Journal of Nutrition*. 2008;7(1):27-30.
8. Amarakoon R. Study on amino acid content in selected varieties of *Pisum sativum* (peas) by ion exchange chromatography. *International Conference on Nutrition and Food Sciences*. 2012;39:211-214.
9. Anyalogbu EA. Human nutrition. In Anyadoh & Ezeji (eds.), *Undergraduate biology I*. Owerri: Readon Publishers Ltd. 2010:49-72.
10. Seligson FH, Mackey LN. Variable predictions of protein quality by chemical score due to amino acid analysis and reference pattern. *Journal of Nutrition*. 1984;114:682-691.
11. AOAC (Association of Official Analytical Chemists), *Official method of Analysis of the AOAC* (W. Horwitz Editor) Eighteenth Edition, Washington, D.C; 2006.
12. Spackman DH, Stein NH, Moore S. Automatic recording apparatus for use in chromatography of amino acid. *Journal of Analytical Chemistry*. 1958;30:1990-1991.
13. FAO/WHO. Protein Quality Evaluation. Report of joint FAO/WHO expert consultation. FAO Food Nutrition paper No. 51. Rome; 1991.
14. Ogunlade I, Olaifa O, Adeniran OA, Ogunlade AO. Effect of domestic processing on the amino acid profile of *Dioscorea rotundata* (White yam). *African Journal of Food science*. 2011;5(1):36-39.
15. Nwosu JN, Ubbaonu CN, Banigo EOI, Uzomah A. The effect of processing on amino acid profile of "Oze" (*Bosquicia angolensis*) seed flour. *Life Science Journal*. 2008;5(4):69-74.
16. Dedeke GA, Owa SO, Olurin KB. Amino acid profile of four earthworm's species from Nigeria. *Agriculture and Biology Journal of North America*. 2010;1(2):97-102.
17. British Pharmacopoeia. Appendix III L. amino acid analysis. British Pharmacopoeia Online; 2012.
Available: <http://bp2012.inforstar.com.cn/Bp2012.aspx?a=display&id=992>.
(Accessed: 01-05-2013).
18. Friedman M. Application of the ninhydrin reaction for analysis of amino acids, peptides, and proteins in agricultural and biomedical sciences. *Journal of Agriculture and Food Chemistry*. 2004;52:385-406.
19. FAO/WHO. Energy and protein requirements. Technical report series no. 522. Geneva, Switzerland: WHO; 1973.
20. Alsmeyer RH, Cunningham AE, Happich ML. Equations to predict PER from amino acid analysis. *Food Technology*. 1974;28:34-38.
21. Adeyeye EI. Effect of cooking and roasting on the amino acid composition of raw groundnut (*Arachis hypogaea*) seeds. *Acta Scientiarum Polonorum, Technologia Alimentaria*. 2010;9(2):201-216.
22. Duncan BO. Multiple range and Multiple F-tests. *Biometrics*. 1955;11:1-42.
23. Olaofe O, Okiribiti BY, Aremu MO. Chemical evaluation of the nutritive value of smooth luffa (*Luffa cylindrica*) seed kernels. *Electronic Journal of Environmental, Agricultural and Food Chemistry*. 2008;7:3444-3452.
24. Aremu MO, Olaofe O, Basu SK, Abdulazeez G, Acharya SN. Processed cranberry bean (*Phaseolus coccineus* L.) seed flour for the African diet. *Canadian Journal of Plant science*. 2010;90:719-728.
25. Aremu MO, Nweze CC, Alade P. Evaluation of protein and amino acid composition of selected spices grown in the middle belt region of Nigeria. *Pakistan Journal of Nutrition*. 2011;10:991-995.
26. Adeola AA. Amino acid composition of tamarind fruit growing wild in Oyo town. *Fountain Journal of Natural and Applied Sciences*. 2013;2(1): 1-5.

27. Etzel MR. Manufacture and use of dairy protein fractions. *The Journal of Nutrition*. 2004;134:996S-1002S.
28. Shimomura Y, Murakani T, Nakai N, Nagasaki M, Harris RA. "Exercise Promotes BCAA Catabolism: Effects of BCAA supplementation on skeletal muscle during exercise." *The Journal of Nutrition*. 2004;1583S-1587S.
29. Combaret L, Dardevet D, Rieu I, Pouch M, Bech D, Taillandier D, Grizard J, Attaix D. A leucine-supplemented diet restores the defective postprandial inhibition of proteasome-dependent proteolysis in aged rats' skeletal muscle. *Journal of Physiology*. 2005;569(2):489-499.
30. Blomstrand E, Eliasson J, Karlsson HK, Köhnke R. Branched-chain amino acids activate key enzymes in protein synthesis after physical exercise. *Journal of Nutrition*. 2006;136(1):269S-273S.
31. Alvares TS, Meirelles CM, Bhambhani YN, Paschoalin VM, Gomes PS. L-Arginine as a potential ergogenic aid in healthy subjects. *Sports Medicine*. 2011;4(3):233-48.
32. Food and Agriculture Organization of the United Nations, FAO. Amino acid content of foods and biological data on proteins. Food Policy and Food Science Service, Nutrition Division, FAO, Rome; 1981.
Available: <http://www.fao.org/docrep/005/ac854t/AC854T23.htm> (Accessed 1/5/2014)
33. El-Mahdy AR, El-Sebaïy LA. Proteolytic activity, amino acid composition and protein quality of germinating fenugreek seeds (*Trigonella foenum graceum* L). *Food Chemistry*. 1985;18:19-23.
34. Alozie Y, Akpanabiatu, MI, Eyong EU, Umoh IB, Alozie G. Amino acid composition of *Dioscorea dumetorum* varieties, *Pakistan Journal of Nutrition*. 2009;8(2):103-105.
35. Stryer L. *Biochemistry*, PWN, Wyd. III; Nauk, Warszawa; 1997.
36. Onimawo IA, Adukwu BI. Proximate Composition and Selected Physicochemical Properties of the Fruits of *Canarium schweifurthii* Linn. *African Journal of Science and Technology*. 2003;4(1):1-4.
37. Oboh G. Effect of blanching on the green leafy vegetables. *Food Science Technology LWT*. 2005;38:513-517.
38. Sefa-Dedeh S, Stanley DW, Voisey PW. Effect of soaking time and cooking conditions on texture and microstructure of cowpeas (*Vigna unguiculata*). *Journal of Food science*. 1978;43:1832-1838.
39. Hotz C, Gibson, RS. Symposium: Food-based approaches to combating micronutrient deficiencies in children of developing countries. Traditional food-processing and preparation practices to enhance the bioavailability of micronutrients in plant-based diets. *Journal of Nutrition*. 2007;137:1097-1100.
40. Yadav SK, Sehgal S. Effect of domestic processing and cooking methods on total, HCl extractable iron and in vitro availability of iron in spinach and amaranth. *Nutrition Health*. 2002;16:113-120.
41. Akinyele BJ, Oloruntoba OS. Comparative studies on *Citrullus vulgaris*, *Citrullus colocynthis* and *Cucumeropsis mannii* for 'Ogiri' production. *British Microbiology Research Journal*. 2013;3(1):1-18.
42. Wei ZY, Wang JS, Cao ZY, Zhang ZY, Li XJ. Effect of high temperature high humidity on contents of protein, amino acids and sugars in wheat grains. *Journal of Chinese Cereals Oils Association*. 2009;24(9):5-11.
43. Gernah DI, Ega BM, Umoh UE. Effect of boiling time on the quality of Zogale: A snack food produced from peanut (*Arachis hypogaea*) cake and boiled *Moringa oleifera* leaves. *African Journal of Food Science*. 2012;6(10):287-293.
44. Dietary Reference intakes (DRIs). Recommended intakes for individuals. Food and Nutrition Board, institute of medicine, National Academics; 2004.

45. Aremu MO, Olonisakin A, Bako DA, Madu PC. Compositional studies and physicochemical characteristics of cashew nut (*Anacardium occidentale*) flour. Pakistan Journal of Nutrition. 2006;5(4):328-333.
46. FAO/WHO/UNU. Energy and Protein Requirements. Technical report series, No. 724, Geneva, Switzerland; 1985.
47. Oluwaniyi OO, Dosumu OO, Awolola GV. Effect of local processing methods (boiling, frying and roasting) on the amino acid composition of four marine fishes commonly consumed in Nigeria. Food Chemistry. 2010;123:1000-1006.
48. Adeyeye EI, Adamu AS. Chemical composition and food properties of *Gymnarchus niloticus* (Trunk fish). Bioscience Biotechnology Research Asia. 2005;3(2):266-272.
49. FAO/WHO. Protein Quality Evaluation. Report of Joint FAO/WHO consultation, Bethesda MD, 4–8 December, 1989. Rome; 1990.
50. Adeyeye EI. The composition of the winged termites, *Macrotermes bellicosus*. Journal of Chemical Society of Nigeria. 2005;30(2):145-9.
51. Krzyściak W. Activity of selected aromatic acids in biological systems. *Acta Biochimica Polonica*. 2011;58:461-466.
52. Friedman M. Nutritional value of proteins from different food sources: A review. Journal of Agriculture Food Chemistry. 1996;44:6-29.
53. Salunkhe DK, Kadam SS. Handbook of world food legumes, nutritional chemistry, processing technology and utilization. Boca Raton, CRC Press Florida; 1989.
54. Oyarekua MA, Eleyinmi AF. Comparative evaluation of the nutritional quality of corn, sorghum and millet ogi prepared by modified traditional technique. Food Agriculture Environment. 2004;2(2):94-99.
55. Deosthale YG, Mohan VS, Rao KV. Varietal deficiencies in protein, lysine and leucine of grain sorghum. Journal of Agricultural and Food Chemistry. 1970;18:644-646.
56. Adeyeye, EI and Aremu, MO. Comparative evaluation of the amino acid profile of the brain and eyes of guinea fowl (*Numida meleagris*) hen. The Open Nutraceuticals Journal. 2010;3:220-226.
57. Adeyeye EI, Asaolu SS, Aluko AO. Amino acid composition of two masticatory nuts (*Cola acuminata* and *Garcinia kola*) and a snack nut (*Anacardium occidentale*). International Journal of Food Science Nutrition. 2007;58(4):241-249.
58. Atmaca G. Antioxidant effects of sulfur containing amino acid. Yonsei Medical Journal. 2004;45:776-788.
59. Emelyn L.(2010). 4 Essential amino acids for hair growth. Available:<http://voices.yahoo.com/4-essential-amino-acids-hair-growth-6522576.html?cat=69>. Accessed: 26-8-2013.
60. Mendoza C. Effect of genetically modified low phytic acid plants on mineral absorption. International Journal Food Science Technology. 2002;37:759-67.
61. FAO. Protein requirements. FAO Nutrition Studies, number 16, FAO, Rome. 1957:52.
62. Bingham S. Nutrition: Better health through good eating. Transworld, ealing. London; 1978.

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