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Effect of Heat Treatment on the Amino Acid Profile of *Plukenetia conophora* Seed Kernel Flours

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Authors' contributions

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

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ABSTRACT

The effect of heat treatment on the amino acid composition of *Plukenetia conophora* (African walnut) seed flours was investigated. The seed kernel of *Plukenetia conophora* (PC) is eaten roasted or cooked mainly as indigenous snacking nut in Nigeria. The sample was divided into four lots. The 1st was used raw, the 2nd, 3rd and 4th boiled in water (99±1°C) for 45, 90, and 135 min and labeled PC_{raw}, PC_{45}, PC_{90}, and PC_{135} respectively. The kernels were found to contain amino acids found naturally in plant protein. Glutamic acid (7.88-18.5 g/100 g protein) and aspartic acid (4.86-9.16 g/100 g protein) were the most abundant non-essential amino acids while the essential amino acid ranges were for leucine (4.50-7.80 g/100 g protein), lysine (3.65-7.09 g/100 g protein) and arginine (3.22-6.12 g/100 protein). In addition to leucine other branched-chain amino acids (Isoleucine and valine) were present in high proportion. Cooking progressively decreased the level of all amino acids. For essential amino acid, the percent decrease ranged from 6.07% for lysinein PC_{45} to 64%

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for isoleucine in PC_{135}. Protein quality parameters such as: predicted protein efficiency ratio, total amino acid, total essential amino acid, total acidic amino acid, total neutral amino acid, total aromatic amino acid and total sulphur amino acid though decreased by cooking were comparable to those of some plant foods and recommended standards. Based on whole hen's egg amino acids, FAO amino acid provisional pattern and Food and Nutrition Board/Institute of Medicine (FNB/IOM) pattern for 1-3-year-old child, the percentage adequacy (Essential amino acid score) of most of the essential amino acids in the samplesat all levels of cooking were high. Despite the decreasein the amino acid contents bycooking, the plant food has the potential for giving high quality protein that can be exploited to enhance protein quality in human nutrition and performance in sports.

Keywords: Plukenetia conophora; heat treatment; amino acid composition; chemical score.

ABBREVIATIONS

Lys, lysine; His, histidine; Arg, arginine; Thr, threonine; Val, valine; Met, methionine; Ile, isoleucine; Leu, leucine; Phe, phenylalanine; Asp, aspartic acid; Ser, serine; Glu, glutamic acid; Pro, proline; Gly, glycine; Ala, alanine; Cys, cysteine.

1. INTRODUCTION

Plukenetia conophora (African walnut) is a climbing and twining tropical plant (Liane) over 30m long. It is of the family Euphorbiaceae and is commonly known in Nigeria as "Ukpa" (Igbo), "Asala" or "Awusa" (Yoruba), "okhue or okwe" (Edo) [1,2]. The fruit is a capsule, 6-10cm long by 3-11cm wide containing sub-globular seeds 2 - 2.5cm long with a thin brown shell resembling the template walnut (Plate1). The seed kernel is eaten raw, roasted or cooked mainly as indigenous snacking nut (masticatory). The consumption of wild fruits and seeds in Nigeria has been reported to be enormous [3,4].

Protein from plant sources remains a major alternative source for the common man since animal protein is scarce and expensive beyond their reach [5]. Some amino acids are regarded as essential for humans because their body is either unable to synthesize them at all or in sufficient quantity from other compounds and must be taken ready-made as part of the diet otherwise they are non-essential [6]. A dietary requirement for protein is a requirement for non-essential amino nitrogen and essential amino acids [7]. Hence, the amino acid composition is the most important factor in defining food protein quality [8-10].

Most processing methods, especially heat processing have detrimental effect on nutrients. Tuleum et al. [11] reported that increase in boiling time significantly decreased the crude protein and most essential amino acid contents of *Mucunautilis.* Earlier, Pisarikova et al. [12], reported that heat processing can cause damage of essential amino acids resulting in decreased contents or transfer into racemic mixture. Other researchers reported either increased or decreased levels of amino acids after processing [5,13-15].

Plate 1. *Plukenetia conophora* (African walnut) seeds

The aim of this research is to determine the effect of heat treatment on the amino acid profile of *Plukenetia conphora* seed kernel flours.

2. MATERIALS AND METHODS

Fresh fruit capsules of *P. conophora* were purchased from a farmer at Ojoto, Idemili South Local Government Area of Anambra State, Nigeria. The capsules were cut with a sharp knife and forty wholesome seeds were collected.

2.1 Sample Processing

The plant samples were washed in several changes of distilled water and divided into four lots. The first lot was used raw and therefore labeled PC_{raw}. Trial processing showed that eating tenderness was obtained by boiling the plant food in water (99±1°C) for 90 min. The 2nd, 3rd and 4th lots were boiled in water (99±1°C) for 45, 90, and 135 min and labelled PC_{45}, PC_{90}, and PC_{135} respectively and the water discarded. The shell of PC_{raw}, PC_{45}, PC_{90} and PC_{135} were cracked with a hammer and the kernels collected, sliced with a knife and dried for 48 hr in an air-circulatory oven (50°C) (Universalwärmeschrank, UNB 100). 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.

2.2 Determination of Amino Acid Concentration

The method of Onyeikeet al. [8] was adapted for this analysis. All the analyses were done in duplicate determinations.

2.2.1 Defatting of sample

Five grammes of the sample flour was weighed into the extraction thimble. Defatting was done for 6hr with 60 ml chloroform/methanol (2:1 v/v) mixture using Soxhlet extraction apparatus as described by AOAC [16].

2.2.2 Acid Hydrolysis of defatted sample

The defatted sample (1.4705 g) was weighed into a glass ampoule and 7ml of 6.0MHCl added [8]. 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 [17]. The glass ampoule was sealed with Bunsen burner flame and incubated in an oven (105°C±5°C) (Universalwärmeschrank, UNB 100) for 22 hr to effect hydrolysis [18]. The ampoule was allowed 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 humins [19,8]. The protein hydrolysate was evapourated to dryness at 40°C under vacuum in a rotary evaporator (BuchiRotavapour, Switzerland) and the residue dissolved with 5ml acetate buffer (pH 2.0), put in plastic specimen bottle and stored in the freezer (–4°C) for further use.

2.2.3 Amino Acid Analysis

With a clean microsyringe (World Precision Instruments®,UK), 10 ml of the hydrolysate was collected and dispensed into the cartridge of Technicon Sequential Multisample (TSM) Amino acid Analyzer (Technicon Instruments Corporation, New York). The analyzer is sodium-based cation-exchange chromatography with post column ninhydrind erivatization [20,21]. The amino acids were separated on the ion-exchange column through a combination of changes in pH and cation strength [22]. 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 [23]. The reaction was monitored at 440 nm and 570 nm wavelengths. The period of analysis was 76 min for each sample. The gas flow rate was 0.58 ml/min at 60°C with reproducibility consistent within ±3%. Norleucine was added as internal standard and a standard mixture of amino acids (Beckman No. 338088, Beckman Coulter, CA) 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/100 g protein from the peak area.

2.2.4 Determination of protein quality parameters

The amino acid score for the essential amino acids was calculated using the FAO/WHO [24] formula as adapted by Onyeike et al. [8]:

$$Aminoacidscore = \frac{Amountofaminoacidinsampleprotein\left(\frac{mg}{g}\right)}{Amountofaminoacidinreferenceproteinpattern\left(\frac{mg}{g}\right)} x100$$

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 Alsmeyeret al. [25], as adapted by Adeyeye [13]:

P-PER = –0.468 + 0.454 (Leu) – 0.105 (Tyr).

3. RESULTS AND DISCUSSION

The essential and non-essential amino acid compositions (g/100 g crude protein) of raw and cooked *P. conophora* seed kernel flours are shown in Table 1 and compared with those of other plant foods in Table 2. The sample at all levels of processing contained the amino acids found naturally in plant protein. Fig. 1 shows the effect of cooking time on the amino acid concentration of the sample represented as percentage changes in the concentrations of essential and non-essential amino acids of the sample when subjected to different cooking time. Table 3 shows the concentrations of total amino acids (TAA), total non-essential amino acids (TNEAA), total essential amino acids (TEAA), total neutral amino acids (TNAA), total acidic amino acid (TAAA), total aromatic amino acids (TArAA), and their percentage values. The predicted protein efficiency ratio, Leu/Ile ratio and their percentage values are also contained in Table 3. The values were affected by the processing method. Tables 4, 5 and 6 show the essential amino acid scores (EAAS) of raw and cooked *P. conophora* seed kernel flours based on whole hen's egg amino acids [26], FAO amino acid provisional pattern [27] and Food and Nutrition Board/Institute of Medicine (FNB/IOM) pattern for one to three year old child [28] respectively. All the patterns indicated reduction in the protein quality of the sample with increasing processing time.

From Table 1, Glu was the most abundant non-essential amino acid in the samples analyzed, with values ranging from 7.88 to 18.5g/100g crude protein followed by Asp which ranged from 4.86 to 9.16g/100 g crude protein. Both amino acids are acidic amino acids. Adeyeye [14] and FAO [26], reporting on groundnut seeds and soybeans respectively (Table 2) also showed Glu and Asp as the most abundant amino acids. Anyalogbu et al. [29] reported Glu as the prominent amino acid in *Canarium schweinfurthii* seed. Earlier, El-Mahdy and El-Sebaiy [30] reported that high concentrations of Glu and Asp were normal for most seeds.

Table 1. Amino acid concentrations (g/100 g protein) of raw and cooked *P. conophora* seed kernel flours

Amino acid	Samples			
	PC_{raw}	PC_{45}	PC_{90}	PC_{135}
Lys*	7.09	6.66	4.08	3.65
His*	3.01	2.29	1.63	1.50
Arg*	6.12	4.60	3.49	3.32
Thr*	3.03	2.66	2.05	1.61
Val*	4.42	3.31	1.92	1.66
Met*	1.12	0.94	0.55	0.44
Ile*	3.51	3.01	1.66	1.26
Leu*	7.80	6.04	5.11	4.50
Phe*	4.14	3.55	2.70	2.20
Asp	9.16	7.92	5.11	4.86
Ser	2.48	1.75	1.24	0.70
Glu	18.50	16.20	8.08	7.88
Pro	3.08	2.65	1.80	1.49
Gly	4.08	3.60	1.92	1.31
Ala	3.94	3.25	2.08	1.78
Cys	3.06	2.90	2.42	2.09

**Essential amino acid, Values are means of duplicate determinations*

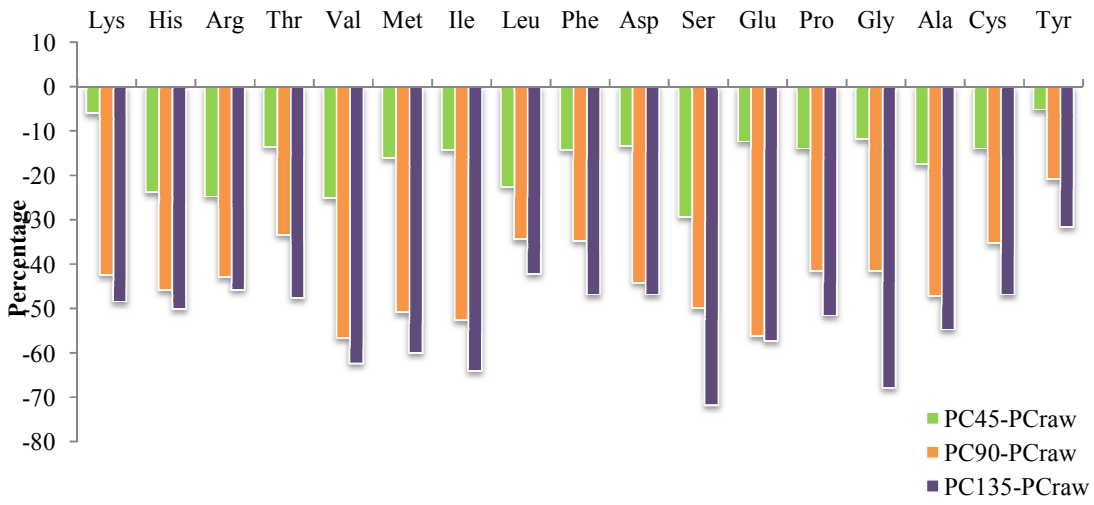

Fig. 1. Effect (%) of cooking time (min) on the amino acid concentration of *P. conophora* seed kernel

Table 2. Amino acid composition of raw and cooked *P. conophora* seed kernel flours compared with those of some plant foods

Amino acid	Samples and other plant foods (g/100 g protein)							
	PC_{raw}	PC_{45}	PC_{90}	PC_{135}	Soybean	Cashew nut	Groundnut seeds	Wheat
Lys	7.09	6.66	4.08	3.65	6.38	3.20	3.71	2.56
His	3.01	2.29	1.63	1.50	2.53	2.40	2.20	2.56
Arg	6.12	4.60	4.49	3.32	7.23	4.40	5.78	4.86
Thr	3.08	2.66	2.05	1.61	3.86	2.50	2.82	2.62
Val	4.42	3.31	1.92	1.66	4.80	3.70	4.41	4.22
Met	1.12	0.94	0.55	0.44	1.24	2.0	0.94	1.63
Ile	3.51	3.01	1.66	1.26	4.54	4.0	3.64	3.37
Leu	7.80	6.04	5.11	4.50	7.78	8.90	7.59	6.31
Phe	4.14	3.55	2.70	2.20	4.94	4.10	4.83	3.95
Asp	9.16	7.92	5.11	4.86	11.7	5.70	9.41	4.95
Ser	2.48	1.75	1.24	0.70	5.12	3.70	3.19	3.92
Glu	18.50	16.20	8.08	7.88	18.70	19.10	14.60	28.60
Pro	3.08	2.65	1.80	1.49	5.49	4.10	3.54	8.99
Gly	4.08	3.60	1.92	1.31	4.18	3.0	3.66	3.86
Ala	3.94	3.25	2.08	1.78	4.26	3.50	3.79	3.37
Cys	1.13	0.99	0.73	0.60	1.33	0.30	1.39	2.18
Tyr	3.06	2.90	2.42	2.09	3.14	3.30	3.27	2.59
TAA	_	_	_	_	97.20	75.80	78.80	90.20
TEAA	_	_	_	_	43.30	33.20	35.90	31.70
%TEAA	_	_	_	_	44.60	43.80	45.60	31.10
Reference	Present study	Present study	Present study	Present study	FAO, [26]	Aremu et al. 2006 [44]	Adeyeye, 2010 [14]	Khan & Eggum, 2006 [45]

The high level of Asp is of importance as it is a precursor of such amino acids as Asn, Thr and Lys [31]. The lowest value for non-essential amino acids occurred in serine (2.48 g/100 g) followed by cysteine (3.06 g/100 g). Leu had the highest value (7.80 g/100 g crude protein) for essential amino acid in all the samples, followed by Lys (7.09 g/100 g crude protein) (Table1). The abundance of Leu in the test samples is of nutritional significance as together with Val (1.66-

4.42 g/100 g protein) and Ile (1.26-3.51 g/100 g protein) it constitutes the branched-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 [32-34]. The plant food also contains a considerable level of Arg (3.32-6.12 g/100 g protein) which is a precursor of nitric oxide (NO). NO promotes vasodilation in active muscle during exercise thereby improving strength and recovery [35]. According to Falavigna [36], increasing the dietary intake of BCAAs to 3.57% was able to improve exercise by 37%. Considering the high content of BCAAs and Arg in the sample, the consumption by the sporting community is highly recommended for improved performance.

Table 3. Concentrations ofessential amino acids parameters, P-PER[a] and Leu/Ile ratio of raw and cooked *P. conophora* seed kernel flour

Quality parameter	PC_{raw}	PC_{45}	PC_{90}	PC_{135}
Total amino acid (TAA)	83.4	74.6	46.6	40.8
Total non-essential amino acid (TNEAA)	43.1	41.6	23.4	20.7
%TNEAA	51.7	55.8	50.2	50.7
Total essential amino acid (TEAA):				
with His	40.3	33.1	23.2	20.1
without His	37.3	30.8	21.6	18.6
% TEAA with His	48.3	44.4	49.8	49.3
% TEAA without His	44.7	41.3	46.4	45.6
Total neutral amino acid (TNAA)	41.8	34.7	24.2	19.6
% TNAA	50.1	46.5	51.9	48.0
Total acidic amino acid (TAAA)	27.7	24.1	13.2	12.7
% TAAA	33.2	32.3	28.3	31.2
Total basic amino Acid (TBAA)	16.2	13.6	9.20	8.47
% TBAA	19.5	18.2	19.7	20.8
Total sulphur amino acid (TSAA)	2.25	1.93	1.28	1.04
% TSAA	2.70	2.59	2.75	2.55
% Cys in TSAA	50.2	51.3	57.0	57.7
Total aromatic amino acid (TArAA)	7.20	6.45	5.12	4.29
% TArAA	8.63	8.65	11.0	10.5
P-PER[a]	2.75	1.97	1.60	1.36
Leu/Ile ratio	2.22	2.01	3.08	3.57
Leu – Ile (difference)	4.29	3.03	3.45	3.24
% Leu-Ile	55.0	50.20	67.50	72.0

[a]Predicted Protein Efficiency Ratio.

Table 4. Essential amino acid scores of raw and cooked *P. conophora* seed kernel flours based on whole hen's egg amino acid[a]

Essential amino acid	Hen's egg (mg amino acid/g Total N)	Chemical score (%)			
		PC_{raw}	PC_{45}	PC_{90}	PC_{135}
Lys	436	101	95	58	52
His	152	123	94	67	61
Thr	320	60	52	40	31
Arg	381	100	75	57	54
Val	428	64	48	28	24
Met	210	33	27	16	13
Met+Cys	362	38	33	22	18
Ile	393	55	48	26	20
Leu	551	88	68	58	51
Phe	358	72	68	58	51
Phe +Tyr	618	73	65	51	43

[a]*Food and Agriculture Organization of the United Nations [26]*

Table 5. Essential amino acid scores of raw and cooked *P. conophora* seed kernel flours based on FAO amino acid provisional pattern[a]

Essential amino acid	FAO provisional amino acid pattern (mg amino acid/g total N)	Chemical score (%)			
		PC_{raw}	PC_{45}	PC_{90}	PC_{135}
Lys	270	164	154	94	84
His	87	216	164	117	108
Thr	180	106	92	71	55
Val	270	102	76	44	38
Met	144	48	40	23	18
Met+Cys	270	52	44	29	24
Ile	270	81	69	38	29
Leu	306	159	123	104	92
Phe	180	143	123	93	76
Phe+Tyr	360	121	108	86	72

[a]*FAO [27]*

Table 6. Essential amino acid scores of raw and cooked *P. conophora* seed kernel flours based on FNB/IOM pattern[a] for 1-to-3-yr-old child

Essential amino acid	FNB/IOM scoring pattern[a] (mg amino acid/g total N)	Chemical score (%)			
		PC_{raw}	PC_{45}	PC_{90}	PC_{135}
Lys	381	139	130	80	71
His	112	167	127	91	83
Thr	168	114	98	76	59
Val	200	138	103	60	51
Met+Cys	156	89	76	51	41
Ile	156	140	120	66	50
Leu	343	141	109	93	81
Phe +Tyr	293	153	137	109	91

[a]*Food and Nutrition Board/Institute of Medicine [28]*

Nitric oxide also plays a major role in male sexual function. It is used by the inner lining of blood vessels to signal the surrounding smooth muscle to relax. This leads to vasodilation (blood vessel dilation), and therefore increased blood flow. Consequently an abundance of nitric oxide can improve circulation and lead to more frequent erections [37]. Studies have shown that when supplementing with L-arginine in doses of 5g or 2.8g daily, men with erectile dysfunction or impotence experienced significant improvements in erectile function [38,39]. With the observed level of Arg (3.32-6.12 g/100 g protein) the plant food holds great potential for application as dietary intervention to the health condition (erectile dysfunction or impotence).

High content of Lys in all the samples is also of nutritional significance since the amino acid is usually limiting in most cereals and legumes [8]. In addition, lysine together with threonine (1.61-3.03 g/100 g protein) is strictly indispensable among the nine indispensable amino acids, since they are not transaminated and their deamination is irreversible [40]. Incidentally, the plant food is commonly consumed as traditional snack in most part of Nigeria where cereals and legumes constitute parts of the staple diet. So, it has the potential for augmenting the daily Lys intake of the consumer.

The values for Met (0.44 - 1.12 g/100 g crude protein), though the lowest in all the samples analyzed was comparable with the values (0.24 - 1.25 g/100 g crude protein) reported for most legumes [31].

The result of this study has shown that Met is the most or first limiting essential amino acid in all the samples. This essential amino acid was also the most limiting in plant foods shown in Table 2.

The concentration of all the amino acids investigated was progressively reduced as the cooking time was extended from 45min (PC_{45}) to 135min (PC_{135}) with the highest effect (71.8%) on serine (at PC_{135}) and least (5.23%) on Tyr (at PC_{45}). Pisarikova et al. [12], working with various amaranth species observed similar effect of heat treatment on amino acid levels. Toleum et al.

[10], on their part reported that increase in boiling time significantly reduced Met, Lys, Ile, Thr, Cys, Ala, and Tyr contents of velvet beans. Based on the shown effect of cooking on the amino acids as reflected by the percentage difference between raw and cooked samples (Fig. 1), Tyr was the most stable and, Ser and Val the most vulnerable amino acids in the samples.

The observed decrease in the amino acid level of the sample with increasing cooking time could be accounted for by the leaching of the nutrient into the cooking water. The leaching may have been facilitated by heat which was reported to increase the solubility of nutrients in the processing water [41]. The decrease, to some extent may be attributed to Millard reaction [42]. Muller and Tobin [43] reported that damage is done by Maillard reactions if the heat treatment causes the reactions to go beyond the deoxy-ketosyl stage. Also according to Adeyeye [14], heat treatment at high temperature causes isomerization of amino acid residues. This involves the deamination reaction and formation of a carbanion which randomly generate L- or D-racemic forms after protonation, and that racemization of the essential amino acid residues of protein reduces its nutritional value to about 50%.

The total amino acid (TAA) of the raw sample (PCraw) and sample cooked for 45 min (PC_{45}) were 83.4 and 74.6 g/100 g crude protein respectively. These were close to the values of 97.2, 75.8, 78.8 and 90.2 g/100 g crude protein obtained for soybeans [26], cashew nut [44], groundnut seeds [14] and wheat [45] respectively (Table 2). Cooking for up to 90 min (PC_{90}) and beyond (PC_{135}) decreased the TAA to between 46.6 and 40.8 g/100 g crude protein. This range is relatively low but is comparable to the value of 49.95 obtained for *Dioscorea rotundata* (white yam) [17]. The TEAA (without His) values of the samples ranged from 20.1 (in PC_{135}) to 40.3g/100g crude protein (in PC_{raw}). This compared favourably with the values of 43.3, 33.2, 35.9 and 31.7 g/100 g crude protein reported for soybean [26], cashew nut [44], groundnut seeds [14] and wheat [45] respectively. The percentage ratio of TEAA to TAA of the samples ranged from 44.4 to 49.8%. This is well above the 39% considered adequate for ideal protein for infants, 26% for children and 11% for adults [46]. The percentage ratio is also comparable to the 50% reported for egg [47].

The TSAA values (1.04-2.25 g/100 g crude protein) obtained for the samples were lower than 5.8g/100g crude protein recommended for infants [46]. The observed % Cys in TSAA ranged from 50.2 to 57.7%. This corroborates the observation of Adeyeye [48] that most vegetable proteins contained more Cys than Met. The values compared favourably with the mean value of 60.9% obtained by Adeyeye [14] for groundnut seeds. It has been shown that dietary Cys has positive effects on mineral absorption, particularly zinc [49]. The aromatic amino acid (ArAA) contents ranged from 4.29 to 7.20g/100g crude protein and are slightly lower than the minimum value of the range (6.8-11.8g/100g crude protein) suggested for ideal infant protein [46].

The P-PER decreased progressively with increasing cooking time (Table 3). Apart from PC_{135}, the P-PER of the plant foods at all levels of processing was higher than 1.50, below which a protein is taken to be of low or poor quality [50]. Heat treatment of amino acid residues of protein generates a mixture of L- and D- racemic forms [14]. The presence of D-isomers reduces the digestibility of the protein because peptide bonds involving D- residues are less easily hydrolyzed *in vivo* than those containing only L-residues and hence, the progressive decrease in P-PER with increasing cooking time.

The Leu/Ile ratio ranged from 2.01 to 3.57. Some research reports have shown that the Leu/Ile balance is important in the regulation of the metabolism of tryptophan and niacin, and hence, some disease processes [51,52]. EAASs (Tables 4, 5 and 6) showed that the protein quality of the sample was decreased by the processing method. Chemical score 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 content of its essential amino acids [8]. Based on Table 4, to correct for amino acid needs i.e. to fulfill the day's needs for all the essential amino acids, from any of the samples, it becomes necessary that 100/33 or 3.03 times as much PC_{raw} protein; 100/27 or 3.70 times as much PC_{45} proteins; 100/16 or 6.25 times as much PC_{90} protein and; 100/13 or 7.69 times as much PC_{135} protein would have to be taken when it is the sole protein source for the individual [53]. Based on egg's protein amino acid pattern (Table 4), His scored highest in PC_{raw} (123%), PC_{90} (95%) and PC_{135} (61%); LYS in PC_{45} (95%).

Generally, high EAAS were obtained for Lys, Arg, Leu, and Phe in PC_{raw} and PC_{45} ranging from 62% (Phe in PC_{45}) to 101% (Lys in PC_{raw}). Highest values of 60% and 64% were obtained for Thr and Val respectively only in the raw sample. Based on FAO provisional amino acid pattern (Table 5), His had the highest scores at all levels of cooking ranging from 108% to 216%. This was followed by Lys, 164% (in PC_{raw}) and 154% (in PC_{45}), and Leu 104% (in PC_{90}) and 92% (in PC_{135}).

On this scale, high EAAS were generally obtained for Lys, His, Leu, and Phe at all levels of cooking. Thr showed high scores up to 90 min of cooking and Val and Ile at PC_{raw} and PC_{45} respectively.

Based on the FNB/IOM pattern (Table 6), the sample, when cooked for not more than 45 min would be able to supply more than the required essential amino acid for the pre-school child (1- to 3-year –old child) except for Met and Thr. When cooked to eating tenderness (90 min), the sample would provide the required essential amino acids for the pre-school child if the limiting Met (51% score) is corrected by taking 100/51 or 1.96 times as much of the protein.

4. CONCLUSION

This study has shown that *Plukenetia conophora* seed kernel contains the amino acids found naturally in plant protein. The high level of Lys in the plant food at all levels of processing is of nutritional importance since the amino acid is usually limiting in most cereals and legumes which incidentally constitute the staple diets of the population that consume the plant food as traditional snack. With the high content of BCAAs and Arg observed in the samples, the consumption could enhance performance in sports. Although the concentrations of both essential and non-essential amino acids were progressively decreased as the time of heat treatment increased from 45 to 135 min, the high values obtained for the protein quality parameters and essential amino acid scoresis indicative that the plant food has the potential for giving high quality protein that could be exploited to enhance protein quality of diets for adults and weaning/complimentary feeding for children.

COMPETING INTERESTS

Authors have declared that no competing interests exist.

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