

Comparability of the Amino Acids Composition of Raw, Roasted and Cooked Dehulled *Treculia africana* Seeds Flour Consumed in Nigeria

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Abstract

The objective of this research was to compare the amino acid qualities of *Treculia africana* when its dehulled seeds were subjected to dry heating and wet heating. Effects of traditional processing (roasting and cooking) on the amino acids (AAs) contents of raw dehulled seeds of *Treculia africana* were investigated. Glutamic acid (Glu) had the highest concentration (g/100g crude protein, cp) in the AAs in roasted dehulled seeds flour (RSDF) (19.6) and cooked dehulled seeds flour (CDF) (20.9), but aspartic acid (Asp) in raw dehulled seeds flour (RDF) (9.91). In the essential amino acid (EAA) group, arginine (Arg) was the most concentrated in RDF (6.95), leucine (Leu) in RSDF (10.2) and CDF (11.7). RDF was better concentrated in eleven and twelve amino acids than roasted and cooked dehulled seeds flour respectively. The total EAA (TEAA) (g/100g cp) were 48.3 (RDF); 39.7 (RSDF) and 40.2 (CDF); total non-essential AA (TNEAA): 43.8 (RDF), 56.9 (RSDF) and 59.0 (CDF); predicted protein efficiency ratio (P-PER₁ and P-PER₂) values were RDF (2.09, 2.23), RSDF (3.89, 3.61) and CDF (5.65, 4.27) respectively; isoelectric point (pI) ranged between 5.13 - 5.51; essential amino acid index (EAAI) values were: RDF (89.3), RSDF (98.2), CDF (98.4) and corresponding biological value (BV) of (86.3), (95.3) and (95.5); lysine/tryptophan (Lys/Trp), methionine/ tryptophan (Met/Trp), Lys/Arg and Asp/Glu ratios ranged between 1.71 - 2.46, 0.618 - 1.61, 0.487 - 0.786 and 0.338 - 1.08 respectively. Lys was the limiting AA in both the RSDF and CDF samples. The linear correlation ($r_{=0.05}$) results showed that there were significant differences between values recorded for total AAs, their quality parameters and the amino acids class groups among the three samples. On the whole, the results showed that RDF was better in 10/18 or 55.6%, RSDF was better in 8/18 or 44.4% while CDF was better in 7/18 or 38.9%. Based on these, raw seeds flour would be best recommended in complementing cereals for weaning foods but less for roasted and cooked seeds flour.

Keywords: Traditional processing; African breadfruit; Amino acids

Abbreviations

RWF: Raw whole seeds flour; RDF: Raw dehulled seeds flour; RTF: raw testa flour; RSWF: Roasted whole seeds flour; RSDF: Roasted dehulled seeds flour; RSTF: Roasted testa flour; CWF: Cooked whole seeds flour; CDF: Cooked dehulled seeds flour; CTF: Cooked testa flour; TAA: Total amino acid; TEAA: Total essential amino acid; TNEAA: Total non-essential amino acid; TNAA: Total neutral amino acid; TAAA: Total acidic amino acid; TBAA: Total basic amino acid; TSAA: Total sulphur amino acid; TArAA: Total aromatic amino acid; P-PER: predicted protein efficiency ratio; pI: Isoelectric point; EAAI: Essential amino acid index; IFE: Index of forecasting efficiency; C_A: Coefficient of alienation; S: Significant; NS: Not significant; CV: Coefficient of variation; SD: Standard deviation; IEC: Ion exchange chromatography; TSM: Technicon sequential multisample; + : Reduction in the amino acid due to roasting of the raw sample; - : Enhancement in the amino acid of roasted and cooked samples; BV: Biological value; TV: Table value, cp: Crude protein.

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Introduction

Proper nutrition is necessary for human beings and for us to have a healthy population that can promote development; the relationship between food, nutrition and health should be reinforced. One of the ways of achieving this is through the exploitation of available local resources, in order to satisfy the needs of the increasing population [1]. Africa and the rest of the developing world are characterized by malnutrition and majority of the population depends solely on carbohydrate which has become their major staple food because it is relatively cheaper than proteinous diets especially of the animal origin. Inadequate protein is a prevalent problem. To this end, there is an urgent need to explore the utilization of plant proteins in the formulation of new food products or in conventional food [2]. One of such plant species that could be exploited for its use and application in food formulation is *Treculia africana* seeds (African breadfruit). It is a woody plant grown around homesteads and outlying fields widely known for its large fruit heads that yield edible seeds. The seeds are eaten as a delicacy in Nigeria (especially in the south-eastern parts of the country). People in this area also derive some income from the sale of the seeds (roasted and raw) in the local markets. Adesina and Adeyeye [3] have reported the seeds to contain appreciable level of nutrients as well as the various levels of the nutrients enhancements due to different processing methods (dehulling, cooking and roasting). It has earlier been reported that the seeds are nutritious if adequately processed [4]. Various methods for food processing and preparations such as dehulling significantly increased protein content and greatly reduced condensed tannin and polyphenol levels in Faba bean [5]. Khamgaonkar *et al.* [6] reported that home practices such as soaking; dehulling and cooking can effectively improve the nutritional value of grains. Dehulling has been reported to improve the amino acids profile of African yam bean (*Sphenostylis stenocarpa*) flour [7] and bambara groundnut flours [8].

The nutritive value of a protein depends primarily on its capacity to satisfy the needs for nitrogen and amino acid requirements and therefore, the logical yardsticks by which protein quality are measured [9]. In spite of this, information on the comparative effects of the different traditional processing methods on the nutritive composition (like amino acids) of *Treculia africana* remains scarce. This study is therefore aimed at elucidating the effects of some selected traditional processing methods (dehulling, roasting and cooking) on the amino acid composition on the raw *Treculia africana* seeds flour. This is because, the knowledge of its nutritive value as well as the processing methods which best preserves the nutrient will encourage its cultivation and consumption.

Materials and Methods

Sample Collection and Treatment

The samples of African breadfruit (*Treculia africana*) seeds were obtained from a local farm in Odo-Ayedun town in Ekiti State, Nigeria. The samples were certified in the Department of Plant Science, Ekiti State University, Ado-Ekiti. The seeds were properly sorted to remove the defected ones. The seeds were dehulled, dried and ground into flour.

Treatment of Samples

A quantity of 450 g of the *Treculia africana* seeds used for the analysis was divided into three groups (about 150 g each for raw, roasted and cooked samples). These forms of samples were prepared following the method described by Adeyeye [10].

Amino Acid Analysis

In each case 1g of each sample was weighed into the extraction thimble and the fat extracted with chloroform/methanol mixture (2:1v/v) using a Soxhlet apparatus [11]. The extraction lasted 5-6h.

In each determination, 30 mg of the defatted sample was weighed into glass ampoules. 7 ml of 6 M HCl were added and oxygen expelled by passing nitrogen gas into the sample. The glass ampoules were sealed with a Bunsen flame and put into an oven at $105 \pm 5^\circ\text{C}$ for 22 h. The ampoule was allowed to cool; the content was filtered to remove the humins. The filtrate was evaporated to dryness at 40°C under vacuum in a rotary evaporator. Each residue was dissolved with 5 ml acetate buffer (pH 2.0) and stored in a plastic specimen bottle and kept in a deep freezer. Amino acid analysis was by ion exchange chromatography (IEC) using the Technicon Sequential Multisample (TSM) Amino Acid Analyzer (Technicon Instruments Corporation, New York). The period of analysis was 76 minutes for each sample. The net height of each peak produced by the chart recorder of the TSM (each representing an amino acid) was measured and calculated.

Determination of Amino Acid Quality Parameters

Determination of amino acid scores: Determination of the amino acid scores was first based on whole hen's egg [12]. In this method, both essential and nonessential amino acids were scored. Secondly, amino acid score was calculated using the following formula [13]:

Amino acid score = amount of amino acid per test protein (mg/g) / amount of amino acid per protein in reference pattern (mg/g).

In this method, Met + Cys and Phe + Tyr were each taken as a unit. Also, only essential amino acids determined were scored. Amino acid score was also calculated based on the composition of the amino acids obtained in the samples compared with the suggested pattern of requirements for pre-school children (2-5 years). Here, Met + Cys and Phe + Tyr were each taken as a unit. Also, only essential amino acids including His were scored.

Determination of the essential amino acid index: The essential amino acid index (EAAI) was calculated by the method of Oser [14].

The two more or less arbitrary assumptions upon which the calculation of an integrated essential amino acid index is based are that the minimum ratio of essential amino acid content relative to that of standard protein is 1%, and the maximum 100%.

The Essential Amino Acid Index (EAA index) may be defined as the geometric mean of "the egg ratios", i.e. the ratios of the essential amino acids in a protein relative to their respective amounts in whole egg protein.

$$EAA = \sqrt[n]{\frac{Lys_p}{Lys_s} \times \frac{Trp_p}{Trp_s} \times \dots \times \frac{His_p}{His_s}}$$

in which the subscript *p* refers to the food protein; *s*, the standard protein (whole egg); and *n*, the number of amino acids (counting pairs such as methionine and cystine as one) entering into the calculation. The simplicity of the computation as conducted logarithmically is the model adopted in this report. (The details of the model is beyond this report.)

Computation of biological value (BV): Computation of biological value (BV) was calculated following the equation of Oser [14] as follows:-

$$BV = 1.09 (EAAI) - 11.7$$

A high degree of correlation between EAA indexes and reported biological values are obtained regardless of whether human milk or egg protein is used as the standard. Mitchell has reported the following regression equation based on 48 such comparisons, with a correlation coefficient (*r*) of +0.948.

$$\text{Biological value (BV)} = 1.0747 (\text{EAA index}) - 13.74 \quad (1)$$

The author's smaller series [Oser (14)] comprised 29 comparisons and yielded the following equation:

$$\text{Biological value (BV)} = 1.1403 (\text{EAA index}) - 8.415 \quad (2)$$

On the basis of the foregoing discussion, indicating that the methods of computation employed in both of these series of data yield essentially the same results, the following equation, weighting the two series of 77 comparisons, is derived:

$$\text{Biological value (BV)} = 1.09 (\text{EAA index}) - 11.73 \quad (3)$$

In our present report, equation 3 was used to estimate the BV levels of the samples.

Determination of the predicted protein efficiency ratio: The predicted protein efficiency ratio (P-PER) was determined using the equations derived by Alsmeyer *et al.* [15], i.e.

$$P\text{-PER}_1 = -0.468 + 0.454 (\text{Leu}) - 0.105 (\text{Tyr}).$$

$$P\text{-PER}_2 = -0.684 + 0.456 (\text{Leu}) - 0.047 (\text{Pro})$$

Other determinations: Determination of the total essential amino acid (TEAA) to the total amino acid (TAA), i.e. (TEAA/TAA); total sulphur amino acid (TSAA); percentage cystine in TSAA (% Cys/TSAA); total aromatic amino acid (TArAA), etc; the Leu/Ile ratios were calculated while the isoelectric point (pI) was calculated using the equation of the form [16]:

$$pI_m = \sum_{i=1}^n IPI X_i$$

Where pI_m is the isoelectric point of the mixture of amino acids, IPI is the isoelectric point of the ith amino acid in the mixture and X_i is the mass or mole fraction of the ith amino acid in the mixture [17].

Computation of Lys/Trp, Lys/Arg [18], Asp/Glu and Met/Trp [19]: The ratios of Lys/Trp, Lys/Arg, Asp/Glu and Met/Trp were computed from the amino acid data results.

Statistical Analysis

Calculations made were the mean, standard deviation (SD) and coefficient of variation in percentage (CV %). The data obtained were also subjected to the determination of linear correlation coefficient (r_{xy}), variance (r_{xy}²), linear regression (R_{xy}) and r_{xy} calculated was compared to r_{xy} table at r_{α=0.05} and at n-2 degree of freedom to see if significant difference occurred among the samples [20]; both the coefficient of alienation (C_A) and index of forecasting efficiency (IFE) [21] were also calculated.

Results and Discussion

Results

Amino acid	RDF	RSDF	CDF	Mean	SD	CV%
Glycine (Gly)	6.45	3.12	3.10	4.22	1.93	45.7
Alanine (Ala)	3.79	8.98	9.07	7.28	3.02	41.5
Serine (Ser)	5.42	6.25	6.20	5.96	0.465	7.81
Proline (Pro)	3.42	7.60	8.66	6.56	2.77	42.2
Valine (Val)	6.09	3.60	3.64	4.44	1.43	32.1
Threonine (Thr)	4.71	3.73	3.28	3.91	0.731	18.7
Isoleucine (Ile)	5.94	3.68	3.86	4.49	1.26	28.0
Leucine (Leu)	6.75	10.2	11.7	9.55	2.54	26.6
Methionine (Met)	1.61	1.76	1.56	1.64	0.104	6.33
Aspartic acid (Asp)	9.91	7.43	7.06	8.13	1.55	19.1
Lysine (Lys)	5.46	1.92	1.88	3.09	2.06	66.6
Glutamic acid (Glu)	9.16	19.6	20.9	16.6	6.44	38.9
Phenylalanine (Phe)	6.08	7.62	7.41	7.04	0.835	11.9
Histidine (His)	2.55	1.89	2.16	2.20	0.332	15.1
Arginine (Arg)	6.95	4.20	3.64	4.93	1.77	35.9
Tyrosine (Tyr)	4.87	2.59	2.35	3.27	1.39	42.5
Cystine (Cys)	0.761	1.78	1.70	1.41	0.567	40.1
Tryptophan (Trp)	2.22	1.09	1.10	1.47	0.650	44.2
Total	92.1	96.6	99.2	96.0	3.59	3.74
Crude protein	16.6	10.4	9.20	12.1	3.97	32.9

Table 1: Amino acid profiles (g/100gcp) of raw whole seeds flour (RDF), roasted whole seeds flour (RSDF) and cooked whole seeds flour (CDF) of *Treculia africana*.

Table 1 presents the results of amino acids (g/100g cp) of raw dehulled seeds flour (RDF), roasted dehulled seeds flour (RSDF) and cooked dehulled seeds flour (CDF) of *Treculia africana*. The Table also included crude protein levels in the samples as follows: 16.6 (RDF), 10.4 (RSDF) and 9.20 (CDF) and a CV% of 32.9.

Amino acid	RDF-RSDF (%)	RDF-CDF (%)
Glycine (Gly)	+3.33(+3.35)	+3.35(+51.9)
Alanine (Ala)	-5.19(-137)	-5.28(-139)
Serine (Ser)	-0.83(-15.3)	-0.78(-14.4)
Proline (Pro)	-4.18(-153)	-5.24(-153)
Valine (Val)	+2.49(+40.9)	+2.45(+40.2)
Threonine (Thr)	+0.98(+20.8)	+1.43(+30.4)
Isoleucine (Ile)	+2.26(+38.0)	+2.08(+35.0)
Leucine (Leu)	-3.45(-51.1)	-4.95(-73.3)
Methionine (Met)	-0.15(-9.32)	+0.05(+3.11)
Aspartic acid (Asp)	+2.48(+25.0)	+2.85(+28.8)
Lysine (Lys)	+3.54(+64.8)	+3.58(+65.6)
Glutamic acid (Glu)	-10.4(-114)	-11.7(-128)
Phenylalanine (Phe)	-1.54(-25.3)	-1.33(-21.9)
Histidine (His)	+0.66(+0.39)	+0.39(+15.3)
Arginine (Arg)	+2.75(+3.31)	+3.31(+47.6)
Tyrosine (Tyr)	+2.28(+2.52)	+2.52(+51.7)
Cystine (Cys)	-1.02(-0.94)	-0.94(-123)
Tryptophan (Trp)	+1.13(+50.9)	+1.12(+50.5)
Total	-4.50(-4.89)	-7.10(-7.71)
Crude protein	+6.20(+37.3)	+7.40(+44.6)

Table 2: Differences in the amino acid profiles (from Table 1) between raw and roasted whole seed (RDF-RSDF), and between raw and cooked whole seeds flours (RDF-CDF) samples of *Treculia africana*.

In Table 2, we have the differences between raw and roasted dehulled seeds (RDF-RSDF), and between raw and cooked dehulled seeds flour (RDF-CDF). The following essential amino acids were reduced by roasting (and cooking) by the following respective percentage values: Val 40.9 (40.2), Thr 20.8 (30.4), Ile 38.0 (35.0), Lys 64.8 (65.6), His 0.39 (15.3), Arg 3.31 (47.6) and Trp 50.9 (50.5). The most affected EAA was Lys followed by Trp. Met was only reduced in the cooked sample with a percent value of 3.11. Among the non-essential amino acids that were reduced by roasting and cooking included: Gly (3.35, 51.9), Asp (25.0, 28.8) and Tyr (2.52, 51.7). On the whole, ten out of the eighteen (10/18 or 55.6%) amino acids were reduced by roasting while eleven (11/18 or 61.1 %) were reduced by cooking. In other words, the raw dehulled seeds flour was better in 10 and 11 amino acids than roasted and cooked dehulled seeds flour respectively.

The quality parameters of the amino acids profile of the samples are depicted in Table 3. The CV% values were generally widely varied between 3.57 - 110. The TEAA content had a value range of 39.7-48.3g/100g cp, with the samples having the values in this order: RDF>CDF>RSDF and corresponding TNEAA value of 43.8-59.0 g/100g cp.

Parameter	RDF	RSDF	CDF	Mean	SD	CV%
TAA	92.1	96.6	99.20	96.0	3.59	3.74
TEAA with His	48.3	39.7	40.2	42.7	4.83	11.3
TEAA without His	45.8	37.8	38.1	40.6	4.53	11.2
TNEAA	43.8	56.9	59.0	53.2	8.24	15.5
TArAA	13.1	11.3	10.9	11.8	1.17	10.0
TAAA	19.1	27.0	28.0	24.7	4.88	19.7
TBAA	15	8.01	7.68	10.2	4.13	40.4
TNAA	55.9	9.98	9.48	25.1	26.7	106
TSAA	2.37	3.54	3.26	3.06	0.611	20.0
%TEAA with His	52.4	41.1	40.5	44.7	6.70	15.0
%TEAA without His	49.7	39.1	38.4	42.4	6.33	14.9
%TNEAA	48.7	58.9	59.5	55.7	6.07	10.9
%TArAA	14.2	11.7	11.0	12.3	1.68	13.7
%TAAA	21.2	28.0	28.2	25.8	3.98	15.4
%TBAA	16.7	8.29	7.74	10.9	5.02	46.0
%TNAA	62.2	10.3	9.56	27.4	30.2	110
%TSAA	2.64	3.67	3.29	3.20	0.521	16.3
Cys in TSAA	0.761	1.78	1.70	1.41	0.567	40.1
% Cys in TSAA	32.1	50.3	52.2	44.9	11.1	24.7
P-PER1	2.09	3.89	5.65	3.88	1.78	45.9
P-PER2	2.23	3.61	4.27	3.37	1.04	30.9
Leu/Ile	1.12	2.77	3.03	2.31	1.04	44.9
Leu-Ile	0.814	6.52	7.84	5.06	3.73	73.8
%Leu-Ile	12.1	63.9	67.0	47.7	30.8	64.7
EAAI	89.3	98.2	98.4	95.30	5.197	5.5
pI	5.51	5.13	5.34	5.33	0.190	3.57
BV	86.3	95.3	95.5	92.4	5.25	5.69
Lys/Trp	2.46	1.76	1.71	1.98	0.419	21.2
Met/Trp	0.618	1.61	1.42	1.22	0.527	43.3
Lys/Arg	0.786	0.487	0.516	0.596	0.165	27.7
Asp/Glu	1.08	0.379	0.338	0.599	0.417	69.6

Table 3: Concentrations of essential, non-essential, aromatic, sulphur, etc. (g/100g cp) of raw whole seeds flour (RDF), roasted whole seeds flour (RSDF) and cooked whole seeds flour (CDF) of *Treculia africana* (dry matter of samples).

Amino acids	RDF	RSDF	CDF	Mean	SD	CV%
Glycine	2.15	1.04	1.03	1.41	0.644	45.8
Alanine	0.70	1.66	1.68	1.35	0.560	41.6
Serine	0.69	0.791	0.79	0.755	0.057	7.50
Proline	0.90	2.00	2.28	1.73	0.729	42.2
Valine	0.81	0.48	0.49	0.592	0.189	32.0
Threonine	0.92	0.731	0.643	0.765	0.142	18.5
Isoleucine	1.10	0.657	0.689	0.815	0.247	30.3
Leucine	0.81	1.23	1.41	1.15	0.308	26.8
Methionine	0.50	0.55	0.488	0.513	0.033	6.41
Aspartic acid	0.93	0.694	0.660	0.761	0.147	19.3
Lysine	0.88	0.31	0.303	0.498	0.331	66.5
Glutamic acid	0.76	1.63	1.74	1.38	0.537	39.0
Phenylalanine	1.19	1.49	1.45	1.38	0.163	11.8
Histidine	1.06	0.788	0.900	0.916	0.137	14.9
Arginine	1.14	0.689	0.597	0.809	0.291	35.9
Tyrosine	1.22	0.648	0.588	0.819	0.349	42.6
Cystine	0.42	0.989	0.944	0.784	0.316	40.3
Tryptophan	1.23	0.606	0.611	0.816	0.359	44.0
Total	0.92	0.967	0.993	0.960	0.037	3.85

Table 4: Amino acid scores of raw whole seeds flour (RDF), roasted whole seeds flour (RSDF) and cooked whole seeds flour (CDF) of *Treculia africana* based on whole hen's egg amino acid scoring pattern.

Amino acid	RDF	RSDF	CDF	Mean	SD	CV%
Leucine	1.02	1.55	1.77	1.45	0.386	26.6
Isoleucine	2.12	1.31	1.38	1.60	0.449	28.0
Lysine	0.94	0.331	0.324	0.532	0.354	66.5
Methionine + Cystine	0.95	1.42	1.30	1.22	0.244	20.0
Phenylalanine + Tyrosine	1.74	1.62	1.55	1.64	0.096	5.87
Tryptophan	2.02	0.991	1.00	1.34	0.592	44.2
Threonine	1.39	1.10	0.965	1.15	0.217	18.9
Valine	1.74	1.03	1.04	1.27	0.407	32.1
Histidine	1.34	1.00	1.14	1.16	0.173	15.0
Total	1.39	1.17	1.20	1.25	0.119	9.52

Table 5: Essential amino acid scores of raw whole seeds flour (RDF), roasted whole seeds flour (RSDF) and cooked whole seeds flour (CDF) of *Treculia africana* based on requirements of pre-school child (2 – 5 years) scoring pattern.

Amino acid	RDF	RSDF	CDF	Mean	SD	CV%
Leucine	0.964	1.46	1.67	1.36	0.363	26.6
Isoleucine	1.49	0.92	0.965	1.13	0.317	28.2
Lysine	0.993	0.349	0.342	0.561	0.374	66.6
Methionine + Cystine	0.677	1.01	0.931	0.873	0.174	19.9
Phenylalanine + Tyrosine	1.83	1.70	1.63	1.72	0.101	5.90
Tryptophan	2.22	1.09	1.10	1.47	0.650	44.2
Threonine	1.18	0.933	0.820	0.978	0.184	18.8
Valine	1.22	0.72	0.728	0.889	0.286	32.2
Total	1.24	1.05	1.07	1.12	0.104	9.32

Table 6: Provisional essential amino acid scores of raw whole seeds flour (RDF), roasted whole seeds flour (RSDF) and cooked whole seeds flour (CDF) of *Treculia africana* based on FAO/WHO (1973) standards.

Amino acids scores based on whole hen’s egg amino acid scoring pattern in Table 4 showed that cystine had the lowest score in the raw dehulled seeds flour (0.42) and lysine in both the roasted and cooked dehulled seeds flour (0.310 and 0.303). Therefore to correct for the amino acids scores from the samples, it is 100/42 or 2.38 times as much as raw dehulled seeds protein, 100/31 or 3.23 times as much roasted dehulled seeds protein and 100/30.3 or 3.30 times as much cooked dehulled seeds protein to be taken (consumed) when each of them serve as sole protein source in the diet. In Table 5, lysine had the lowest score based on the requirements of pre-school child (2- 5 years) among all the samples with values of 0.94 (RDF), 0.331 (RSDF) and 0.324 (CDF) and would need correction of 100/94 or 1.06, 100/33.1 or 3.02 and 100/32.4 or 3.09 times as much protein in each if they were to serve as sole protein source in diets. The provisional essential amino acid scores in Table 6 showed that Met + Cys had the least score in RDF (0.677) and would require a correction of 100/67.7 or 1.48 times as much protein if it is to serve as sole source of protein in the diet. Lys was the least score in RSDF (0.349) and CDF (0.342), the AA would be corrected for with a value of 2.87 and 2.93 respectively.

Class	RDF	RSDF	CDF	Mean	SD	CV%
I [with aliphatic side chains (hydrogen and carbons) = Gly, Ala, Val, Leu, Ile]	29.12 (30.7%)	29.58 (29.9%)	31.37 (30.9%)	30.0	1.19	3.96
II [with side chains containing hydroxylic (OH) groups = Ser, Thr]	10.13 (10.7%)	9.980 (10.1%)	9.480 (9.70%)	9.86	0.340	3.45
III [with side chains containing sulphur atoms = Cys, Met]	2.371 (2.50%)	3.540 (3.58%)	3.260 (3.21%)	3.06	0.610	20.0
IV [with side chains containing acidic groups or their amides = Asp, Glu]	19.07 (20.1%)	27.03 (27.3%)	27.96 (27.6%)	24.7	4.89	19.8
V [with side chains containing basic groups = Arg, Lys, His]	14.96 (15.8%)	8.010 (8.10%)	7.680 (7.57%)	10.2	4.11	40.2
VI [containing aromatic rings = His, Phe, Tyr, Trp]	15.72 (16.6%)	13.19 (13.3%)	13.02 (12.8%)	14.0	1.51	10.8
VII [imino acids = Pro]	3.42 (3.61%)	7.60 (7.68%)	8.66 (8.54%)	6.56	2.77	42.2
Total	94.8 (100%)	98.9 (100%)	101 (100%)	98.4	3.34	3.40

Table 7: Amino acid groups of raw whole seeds flour (RDF), roasted whole seeds flour (RSDF) and cooked whole seeds flour (CDF) of *Treculia africana*.

Amino acid composition	Samples (Factor A)			Factor B means
	RDF	RSDF	CDF	
Total essential amino acid	48.3	39.7	40.2	42.7
Total non-essential amino acid	43.8	56.9	59.0	53.2
Factor A means	46.1	48.3	49.6	48.0

Table 8: Summary of the amino acid profiles into Factors A and B.

Tables	Groups compared	r_{xy}	r_{xy}^2	R_{xy}	C_A (%)	IFE (%)	TV	Remark
Table 1	RDF/RSDF	0.98341	0.9671	0.96528	3.29	96.7	0.4716	S
	RDF/CDF	0.97967	0.95795	0.95712	4.20	95.8	0.4716	S
	RSDF/CDF	0.999635	0.99927	0.99923	2.70	97.3	0.4716	S
Table 3	RDF/RSDF	0.821996	0.67568	0.66367	32.4	67.6	0.3685	S
	RDF/CDF	0.811493	0.65852	0.64587	34.1	65.9	0.3685	S
	RSDF/CDF	0.999586	0.99917	0.99914	2.90	97.1	0.3685	S
Table 4	RDF/RSDF	0.041814	0.00175	-0.05697	99.8	0.200	0.4853	NS
	RDF/CDF	0.051563	0.00266	-0.05601	99.7	0.300	0.4853	NS
	RSDF/CDF	0.989712	0.97953	0.97833	14.3	85.7	0.4853	NS
Table 5	RDF/RSDF	0.17436	0.0304	-0.0908	97.0	3.00	0.7143	NS
	RDF/CDF	0.158747	0.0252	-0.09665	74.8	25.2	0.7143	NS
	RSDF/CDF	0.95943	0.92051	0.91057	28.2	71.8	0.7143	NS
Table 6	RDF/RSDF	0.340695	0.11607	-0.0102	88.4	11.6	0.7450	NS
	RDF/CDF	0.30092	0.09055	-0.03937	69.9	30.1	0.7450	NS
	RSDF/CDF	0.975332	0.95127	0.94431	22.1	77.9	0.7450	NS
Table 7	RDF/RSDF	0.990028	0.98016	0.97685	1.00	99.0	0.8236	S
	RDF/CDF	0.988432	0.977	0.97316	1.20	98.8	0.8236	S
	RSDF/CDF	0.999774	0.99955	0.99947	2.10	97.9	0.8236	S

Table 9: Statistical (linear correlation) summary of data in Tables 1, 3, 5, 6 and 7.

The amino acid groups into classes I-VII is shown in Table 7. The general trend in class concentration of AA could be seen as follows: RDF, I>IV>VI>V>II>VII>III, RSDF, I>IV>VI>II>V>VII>III and CDF, I>IV>VI>II>VII>V>III. This meant that the most concentrated AA was the amino acid with aliphatic side chains (RDF: 29.12 g/100g cp, RSDF: 29.58 g/100g cp and CDF: 31.37 g/100g cp) whereas the least concentrated was AA with side chains containing sulphur atoms (RDF, 2.37 g/100g; RSDF, 3.54 g/100g cp and CDF, 3.26 g/100g cp).

The amino acid profiles (g/100g cp) for the samples shown in Table 8 as distributed into Factors A and B means gave a terminal value of 48.0. This was slightly higher than the factor B means of 42.7 (TEAA) and slightly less than 53.2 (TNEAA).

Table 9 depicts the statistical summary of the data in Tables 1, 3-7. The r_{xy} levels were mostly positively high for Tables 1, 3 and 7 (0.811493 - 0.990028) but low for data from Tables 4, 5 and 6 (0.051563 - 0.340695) with corresponding R_{xy} values of 0.645874 - 0.976848 and -0.010202 to -0.09665 respectively.

Discussion

The protein content in legume seeds is governed by the genotype and environmental conditions under which they are grown [22]. It has been shown that the proteins of legumes are mainly deficient in sulphur-containing amino acids and tryptophan, but are rich in lysine, in which cereals are relatively deficient [23- 25]. In the raw dehulled seeds flour, aspartic acid (Asp) had the highest concentration (9.91 g/100g cp), while glutamic acid (Glu) constituted the most concentrated amino acid in the roasted and cooked dehulled seed flours (19.6 and 20.9 g/100g cp respectively). These values are higher than the values reported for raw, roasted and cooked testa of *T. africana* by Adeyeye and Adesina [26], showing that these amino acids may be more concentrated in the dehulled seeds. The levels of aspartic acid recorded in the present report agreed well with the values reported for varieties of African yam bean flour (whole seeds) [27]. However the levels of glutamic acid (Glu) in the present report were comparably higher than the values reported for six varieties of African yam bean flour [7], roasted and cooked testa of African breadfruits seeds flour [26], roasted cocoa, cocoa nibs and cocoa shell [28]. In general, legume protein exhibits a wide range of variations in their essential amino acid composition.

Legume proteins are characterized by high content of polar amino acids with opposite charges (aspartic and glutamic acid on one side and lysine and arginine on the other side) [29]. Therefore it is reasonable to suggest that these amino acids are involved in the association-dissociation phenomena of protein sub-units that are the basis of the solubility properties of legume oligomeric proteins [30]. Among the essential amino acids (EAA) in the raw dehulled seeds flour (RDF), arginine had the highest concentration with a value of 6.95 g/100g whilst in both the roasted and cooked dehulled seeds flour (RSDF and CDF), leucine recorded the highest concentration (10.2 and 11.7 g/100g cp, respectively). These levels were comparably higher than what was reported for roasted cocoa, cocoa nibs and cocoa shell [28], hulled African yam bean flour [7] and *Sesamum indicum* and *Balanite aegytiaca* leaves [31].

Generally, the reduction in the amino acids level observed during processing might apparently be due to the Maillard reactions during which damage is said to be done by heat treatment that causes the reaction to go beyond the deoxy-ketosyl stage. Large amounts of Lys and Arg were destroyed and to a lesser extent, Trp and His [32]. Severe heat treatments at high temperatures cause isomerization of amino acid residues. This involves the deamination reaction and formation of carbanion, which after protonation leads randomly to L or D forms. When the reaction is complete, a racemic mixture of the amino acids is obtained. Since most D-amino acids have no nutritional value, racemization of the residue of an essential amino acid results, thus reducing its nutritional value to about 50% [32, 33].

The values of TNEAA (43.8 - 59.0 g/100g cp) (Table 3) are highly comparable to the values reported for roasted cocoa, cocoa nibs and shells [28]. The % TEAA of 40.5-52.4 were all above the 39% considered adequate for ideal protein food for infants, 26% for children and 11% for adults [13]. The TEAA in the present report compared favourably well with the levels in pigeon pea (45.2 g/100g) [34], pumpkin seed (45.2 g/100g) [35], cowpea (42.6 g/100g) [36]. The TSAA values of 2.37-3.54 g/100g cp in the present report could only satisfy between 40.9-61.0 % of the 5.80 g/100g cp recommended for infants [13]. The TArAA range obtained in the present report of 10.9-13.1 g/100g cp would be sufficient to meet the TArAA range suggested for ideal infant protein (6.8-11.8 g/100g cp).

Although, it is known that cystine can spare part of the requirement of methionine, FAO/WHO/UNU [37] does not give any indication of the proportion of total sulphur amino acids that can be met by cystine. For rat, chick and pig, the proportion is about 50% [38]. It has also been reported that most animal proteins are low in cystine, in contrast, many vegetable proteins, especially the legumes, contain substantially more cystine than methionine. Thus, for animal protein, cystine is unlikely to contribute more than 50% of the total sulphur amino acids [38]. However, in the present report, except in RDF where the value was 32.1%, the roasted and cooked dehulled seeds flour (RSDF and CDF) had percent values of 50.3 and 52.2 respectively for % Cys in TSAA. Logically, it is correct to say that RDF sample did not behave much like plant proteins as RSDF and CDF do under the present conditions.

The predicted protein efficiency ratio (P-PER₁, and P-PER₂) were: RDF (2.09, 2.23), RSDF (3.89, 3.61) and CDF (5.65, 4.27). These values were comparably higher than the ratios reported for roasted cocoa, cocoa nibs and shells [28], *Cucurbita maxima* and *Amaranthus viridis* [39]. Muller and Tobin [32] reported that the experimentally determined PER usually ranged from 0.0 for a very poor protein to a maximum possible of just over 4.0. These present results showed that our samples would be readily utilized in the body. The Leu/Ile values in the present report (RDF: 1.12, RSDF: 2.28 and CDF: 3.03) were in agreement with the ideal Leu/Ile of 2.36 given by FAO/WHO [38] except CDF which fell slightly above the literature value.

The isoelectric point (pI) is the minimum pH at which the protein becomes soluble, thus, in preparing the isolate of protein of any given sample, the pI becomes highly important. From the present report the pI ranged between 5.13-5.51, showing that the minimum solubility pH would be just above 5.0.

The high content of lysine, arginine, aspartic and glutamic acid ratios, i.e Lys/Arg and Asp/Glu acids in the bean proteins would represent the condition for the mechanism (association-dissociation phenomena of protein sub-units that are at the basis of the solubility properties of legume oligomeric proteins) of interaction to occur effectively [19]. Such a mechanism might be common among legumes protein, as all of these are characterized by very high amount of both acid and basic amino acids [29]. Mosses [40] stated that cereal grains and legumes or oilseeds remain by far the predominant source of protein used for human food and for animal feed. The levels of Lys/Arg and Asp/Glu in the present report were: RDF (0.786, 1.08), RSDF (0.457, 0.379) and CDF (0.516, 0.338).

The essential amino acids index (EAAI) ranged from 89.3-98.4 with a CV% of 5.50. The EAAI is used as a rapid tool to evaluate food formulation for protein quality, although it does not account for differences in protein quality due to various processing methods or certain chemical reactions [41]. Albanese [18] had also reported that EAAI is essential for the determination of biological value (BV) of a protein. The EAAI in the present report compared favourably with the EAAI reported for *Pandalus borealis* [42]. The BV levels in the present report of 89.3-98.4 showed the high quality of protein in the raw and processed dehulled seeds of *Treculia africana*.

The ratios of Lys/Trp and Met/Trp in the current report were: 1.71-2.46 and 0.018-1.61 respectively. The values were however much lower than the values reported for *Pandalus borealis* body parts [42] and mammalian tissues recorded by Mitchell [43].

Amino acid groups in Table 7 were comparably higher than those reported for roasted cocoa (21.9 g/100g cp), cocoa nibs (23.9 g/100g) and cocoa shell (12.2 g/100g) [28].

Table 9 is basically a statistical comparative analysis between raw dehulled seeds flour/roasted dehulled seeds flour (RDF/RSDF), raw dehulled seeds flour/cooked dehulled seeds flour (RDF/CDF) and between roasted dehulled seed flour/cooked dehulled seed flour (RSDF/CDF). For example, we can take results from Table 1. The RDF/RSDF (row 1 of Table 9 from Table 1) showed the r_{xy} as a high positive value (0.98341), r_{xy}^2 (coefficient of determination) of 0.9671, and R_{xy} of 0.96528. This meant that for every one unit rise in the raw dehulled seeds amino acid value, there was a corresponding increase of 0.96528 in the roasted dehulled seeds. For the coefficient of alienation (C_A), the value of 3.29 was low whereas the index of forecasting efficiency (IFE) was high at 96.7 signifying that the forecasting efficiency of relationship was high. The index of forecasting efficiency is actually the reduction in the error of forecasting efficiency, the higher the IFE, the lower is the error in the forecasting efficiency. For RDF/CDF, all these parameters were high: r_{xy} , r_{xy}^2 and R_{xy} , C_A

was low. For C_A of 1.00 in Table 7 row 1, it made the IFE very high giving a reduction of 99.0 % in the error of prediction of relationship between raw dehulled seeds and roasted dehulled seeds amino acid profiles. The R_{xy} value of -0.05697 in Table 4 row 1, showed that for every one unit increase in the amino acid level of raw dehulled *Treculia africana* seeds, there was a reduction of 0.05697 in the roasted dehulled seeds. Similar trend of explanation holds for the rest of the groups and data.

All the r_{xy} values were significant except for values from Tables 4, 5 and 6 at $r_{=0.05}$ and n-2 degree of freedom. The coefficient of alienation (C_A) ranged from 1.00 -34.1 % (for data from Tables 1, 3 and 7) and from 14.3 - 99.8% (for data from Tables 4, 5 and 6) whereas the corresponding index of forecasting efficiency (IFE) ranged between 65.9 - 99.0% (for data from Table 1, 3 and 7) and 0.20 - 85.7 (for data from Tables 4, 5 and 6).

Conclusion

This study has revealed that the dehulled seeds (raw, roasted and cooked) of *Treculia africana* contained useful levels of amino acids that are good for human nutrition. On the whole, the results showed that roasted dehulled seeds flour was better in 8/18 or 44.4% while cooked dehulled seeds flour was better in 7/18 or 38.9%. In all the ten important quality parameters of protein (P-PER₁, P-PER₂, Leu/Ile, EAAI, pI, BV, Lys/Trp, Met/Trp, Lys/Arg and Asp/Glu), the trend was that RDF (better in 4 or 40 % than the RSDF and CDF), RSDF (better in 6 or 60 % than RDF), CDF (better in 6 or 60% than the RDF) also the CDF was better in 7 or 70% of the parameters than in the roasted sample, i.e. CDF>RSDF>RDF. The statistical analysis (linear correlation) showed that significant differences existed between the amino acids profiles, quality parameters and amino acid groups whereas there existed no significant differences at $r_{=0.05}$ among the amino acid scores (based on all the patterns used). Therefore the partial replacement of animal or human foods with such sample *Treculia africana* or its consumption (mostly in the roasted or cooked form) could improve the nutritional status of amino acids in the diets of people in developing countries.

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